





(11) EP 0 702 082 A1

(12)

EUROPEAN PATENT APPLICATION

01 P E (4

(43) Date of publication:20.03.1996 Bulletin 1996/12

(51) Int. Cl.⁶: **C12N 15/13**, C07K 16/42

APR 1 5 2002

(21) Application number: 94115683.8

(22) Date of filing: 05.10.1994

(84) Designated Contracting States:

AT BE CH DE DK ES FR GB GR IE IT LI LU MC NL
PT SE

(30) Priority: 06.10.1993 JP 272950/93

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(54) Amino acid sequences of anti-idiotypic antibodies against anti-cancer human monoclonal antibody, and dna base sequences encoding those sequences

(57) Amino acid sequences of the H chain and L chain variable regions of mouse monoclonal antibodies Idio 3, Idio 17, Idio 20, Idio 27 and Idio 33 against idiotypes of a cancer cell antigen-specific human immunoglobulin CLN/IgG produced by a human/human fused cell strain CLN/SUZ H11, and base sequences of the genes of the variable regions are disclosed.

The above amino acid sequences and the base sequences are useful in medical and pharmaceutical fields such as prophylaxis, treatment and/or diagnosis of human diseases, and/or in pharmacological and/or biochemical fields, etc. such as biochemical reagents, and reagents for purification of biomacromolecules.

Description

Detailed Description of the Invention

This invention relates to the structure of the variable regions of mouse immunoglobulins against idiotypes of an antigen-specific human immunoglobulin, useful in wide fields, for example in pharmaceutical fields such as prophylaxis, treatment and/or diagnosis of human diseases, and/or in pharmacological and/or biochemical fields such as biochemical reagents and reagents for purification of biomacromolecules.

More detailedly, this invention relates to the amino acid sequences of the H chain and L chain variable regions of mouse immunoglobulins against idiotypes of a cancer cell antigen-specific human immunoglobulin produced by a human/human fused cell strain CLN/SUZ H11 from a B cell of a patient carrying human cervical carcinoma and a human lymphoblastoid cell strain, and relates to the base sequences of the genes of the variable regions.

Since the development of the technique of formation of monoclonal antibodies by cell fusion or immortalization of cells, many useful antibodies have been obtained using mainly mice. Among them, monoclonal antibodies against malignant tumor cells are utilized not only for fundamental researches such as analyses of tumor antigens, but in serum diagnoses, image diagnoses of tumors using labeled antibodies, and have extremely high utilization value. Particularly, human-derived anti-cancer monoclonal antibodies are expected as ideal antibodies in the clinical field, since they have only faint or no side effects.

In such circumstances, one of the present inventors, as disclosed detailedly in Japanese Laid-Open Patent Publication No. 201994/1983 (= U. S. Patent No. 5,286,647; EP-A-839,02157.3), Japanese Laid-Open Patent Publication No. 135898/1984 and Japanese Laid-Open Patent Publication No. 137497/1984, established a cell strain CLN/SUS H11 (ATCC No. HB 8307) which produces a human monoclonal antibody having a high reactivity with human cancer cells. Interesting findings are obtained about the antibody (named CLN-IgG) produced by this cell strain, that the antibody class is IgG; the isotypes are γ 1 type and κ type; and the antibody binds to a cancer antigen immunohistologically existing on the surface of the cancer cells and moreover inhibits proliferation of the cancer cells. At present, the whole amino acid sequence and DNA base sequence of the antibody are clarified (Japanese Laid-Open Patent Publication No. 346792/1992 = WO 92/20799).

On the other hand, since Jerne put forward the so-called network theory, various researches have been made on the structure of the variable regions of antibodies. An antibody binds to an antigen at its variable region (antigen combining site). Therefore, the variable regions of antibodies have various three-dimensional-like structures in accordance with the structures of the antigenic determinants on the surfaces of antigens to be recognized. Thus, an antibody itself can be considered to be an antigen, and in the case, the structures of the variable regions of the antibody are called idiotypes, and antibodies against the idiotypes of the antibody are called anti-idiotypic antibodies. The structure corresponding to an antigenic determinant is called an idiotope. An idiotype can be thought to be an aggregate of idiotypes. It was reported that among anti-idiotypic antibodies (Ab2) against an antibody (Ab1) exist antibodies which competitively inhibit binding of Ab1 to an antigen and have idiotopes analogous to antigens recognized by the antibodies, i.e. antibodies having structures as so-called internal images of the antigen.

In view of the above findings, anti-idiotypic antibodies are expected to be utilized for the purpose of treatment and/or diagnosis of cancers.

For example, as for the purpose of cancer treatment, a vaccine therapy using an anti-idiotypic antibody as an antigen is made possible. It is generally difficult to get cancer antigens in large amounts, and it is restricted from a safety aspect and an ethical aspect to directly immunize human beings with cancer cells as antigens. Therefore, these problems can be avoided by performing immunization with an anti-idiotypic antibody in place of an antigen.

In a diagnostic aspect, anti-idiotypic antibodies can be utilized to examine the state of immune reactions against cancer cells. Specifically, it serves for early detection of cancers, judgment of therapeutic effects to detect or determine one's antibodies against cancer antigens existing in the blood or humor of cancer patients.

Under such technical background, problems as stated below are underlying to be solved.

1) When anti-idiotypic antibodies are utilized as vaccines or diagnostic drugs, it is necessary to provide these antibodies in large amounts and stably. 2) There is a possibility to give more powerful vaccines or diagnostic drugs abounding in functionality by altering or modifying the antibodies.

A method by gene manipulation is considered as a means for solving the above problems, i.e. a means for realizing improvement of production amount of the antibodies and elevation or modification of the activities of the antibodies.

For example, in the case of the problem of 1), it can be considered to solve the problem by cloning such an antibody gene, introducing the gene into host cells such as animal cells or <u>Escherichia coli</u>, expressing the antibody gene to give a large amount of the antibody, and in the case of the problem of 2), it can be considered to alter such an antibody so as to have stronger immunogenicity by artificially changing the antibody gene, or to design an antibody molecule having a higher vaccinal activity by adding a function which the antibody does not inherently have, for example an enzymatic activity, an immunity induction activity or the like to the antibody molecule or a fragment thereof.

For accomplishment of these purposes, separation of anti-idiotypic antibody genes, and clarification of their structures are necessary. However, there has not so far been known anything at all about the structures of L chains and H chains constituting anti-idiotypic antibodies against idiotypes of CLN-IgG, and the gene structures of the variable regions having a function to specifically bind to idiotopes of CLN-IgG.

Thus the main object of this invention is to clarify the gene structures of the L chains and the H chains of anti-CLN-IgG idiotype antibodies.

The present inventors have succeeded in creating hybridomas producing, respectively, five kinds of mouse anti-CLN-lgG idiotype antibodies (ldio 3, ldio 17, ldio 20, ldio 27 and ldio 33) having $\gamma 1$ and κ isotypes against the idiotypes of CLN-lgG; have separated, from the hybridomas, cDNAs encoding the L chains and H chains of the anti-idiotypic antibodies, respectively; have clarified their DNA base sequences; have determined, based on these sequences, th amino acid sequences of the L chains and H chains of the antibodies, respectively; and have completed this invention.

Thus, according to this invention are provided an immunoglobulin H chain variable region fragment which contains a hypervariable region CDR1 having an amino acid sequence selected from

(1) Ser Tyr Trp Met His;
Asp Tyr Tyr Met Asn; and
Asn Tyr Trp Met Gln.

a hypervariable region CDR2 having an amino acid sequence selected from

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Asp Ile Tyr Pro Glý Asn Ser
Asp Ile Ser Tyr Ser Gln Asn
Phe Lys Asp;
Phe Ile Arg Asn Lys Ala
Asn Leu Tyr Thr Thr Asp
Tyr Ser Ala Ser Val Lys
Gly;
Phe Ile Arg Asn Lys Ala
Asn Tyr Tyr Thr Thr Glu
Tyr Ser Ala Ser Val Lys
Gly; and
Ala Ile Tyr Pro Gly Asp
Gly Asp Thr Arg Tyr Thr
Glu Lys Phe Lys Gly,

and a hypervariable region CDRs naving an amino acid sequence selected from

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(3) Glu Glu Tyr Asp Tyr Asp
Thr Leu Asp Tyr;
Asp Arg Gly Gly Arg Asp
Trp Tyr Phe Asp Val;
Asp Gly Phe Leu Arg Asp
Trp Tyr Phe Asp Val; and
Ser Gly Tyr Tyr Gly Ser
Phe Val Gly Phe Ala Tyr;

and DNA and RNA fragments encoding the immunoglobulin H chain variable region fragment.

According to this invention are further provided an immunoglobulin L chain fragment which contains a hypervariable region CDR1 having an amino acid sequence selected from

Gln Leu His Leu Ala Ile Val
Tyr Met His;
Tyr Arg Ala Ser Lys Ser Val
Ser Thr Ser Gly Tyr Ser Tyr
Met His;
Lys Ala Ser Gln Asp Val Asn
Thr Ala Val Ala; and
Lys Ala Ser Gln Asp Val Thr
Thr Asp Val Ala;

40 a hypervariable region CDR2 having an amino acid sequence selected from

(2) Leu Val Ser Asn Leu Glu Ser; Leu Val Ser Asn Leu Asp Ser; and Ser Ala Ser Tyr Arg Tyr Thr,



and a hypervariable region CDR3 having an amino acid sequence selected from

(3) Gln His Ile Arg Val Ala Tyr
Thr;
Gln His Ile Arg Gly Ala Tyr
Thr;
Gln His Ile Glu Gly Ala Tyr
Thr;
Gln Gln His Tyr Ser Pro Pro
Leu Thr; and
Gln Gln His Tyr Ser Thr Ala
Trp Thr;

and DNA and RNA fragments encoding the immunoglobulin L chain variable region fragment.

In this invention, cytoplasmic RNAs were prepared from the five mouse hybridomas, respectively; the RNAs were converted to cDNAs by a reverse transcriptase; the antibody genes were amplified using these cDNAs as templates and using the PCR method; the amplified DNA fragments were integrated into plasmids and cloned; the base sequences of the insertion DNAs of the plasmids purified from <u>Escherichia coli</u> clones isolated were determined, and the amino acid sequences were determined based on the base sequences. These steps are further detailedly described below.

[1] Isolation of cytoplasmic RNAs

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Each mouse hybridoma is cultured and proliferated in a culture medium, e.g. and RDF or RPMI 1640 medium, containing 5% fetal bovine serum under a suitable condition, e.g. under a condition of 37°C and a carbon dioxide concentration of 5%; the resultant cells are collected by centrifugation; and the cytoplasmic RNA is extracted from the cells by a conventional method, e.g. a method disclosed in 7.12 of Molecular Cloning (2nd edition, edited by Sambrook et al., Cold Spring Harbor Laboratory Press 1989). The resultant cytoplasmic RNA can further be utilized as a template for cDNA synthesis. Specifically in this invention, the cytoplasmic RNAs were extracted from mouse hybridomas No. 3, No. 17, No. 20, No. 27 and No. 33, and provided for synthesis of cDNAs.

[2] Synthesis of cDNAs

Using a cytoplasmic RNA obtained in the step of [1] as a template, a single-strand DNA complementary to the mRNA is synthesized in the presence of dATP, dGTP, dTTP and dCTP using, as a primer, an oligo dT corresponding to a poly A, or a synthetic nucleotide having a random sequence, and a reverse transcriptase. In the specific operations in the invention, cDNAs were synthesized using the cytoplasmic RNAs obtained in the step of [1] as templates and a random hexamer as a primer, respectively, and provided for the step of amplification of the antibody genes.

45 [3] Amplification of antibody genes by PCR

PCR reaction is performed in the presence of dATP, dGTR, dTTP, dCTP and Taq polymerase using as a template a single-strand cDNA obtained in the step of [2] and as a primer a sequence of the antibody gene (e.g., a sequence encoding a constant region, a variable region or a leader region of the antibody gene) to amplify the antibody gene. Suitably in the invention, the antibody genes were amplified using as templates the single-strand cDNAs obtained in the step of [2] and using synthetic DNA oligomers corresponding to the sequences of the leader regions and variable regions of the L chains and H chains of the antibodies, respectively.

[4] Cloning of PCR-amplified DNA fragments

A PCR-amplified DNA fragment obtained in the step of [3] is, directly or after treatment with restriction enzyme(s), ligated into one of various vectors, for example plasmid vectors such as pUC 18, pCR1000 and pCR™, phage vectors such as M 13 phage, and phagemid vectors such as pUC 118 and pBluescrpt SK* to prepare a vector containing the insertion fragment. Then, <u>Escherichia coli</u> is transformed with the vector, and a colony of the <u>Escherichia coli</u> containing

the targeted antibody gene fragment is obtained. The purified vector recovered from the <u>Escherichia coli</u> is provided as a sample for determination of the DNA base sequence. In the specific operations in the invention, the PCR-amplified DNA fragments obtained in the step of [3] were directly ligated, respectively, into pCR1000 and pCR™ plasmid vector; an <u>Escherichia coli</u> INVαF' was transformed with each of the resultant plasmids; and the plasmids were purified from the resultant <u>Escherichia coli</u> colonies, respectively.

[5] Determination of the base sequences and amino acid sequences of the DNAs

The base sequence of the DNA at the insertion site in a plasmid obtained in the step of [4] can be determined using the Maxam-Gilbert method or the Sanger method. In the invention, the pCR1000 or pCR™ plasmid vectors containing the insertion fragments were purified, respectively; their base sequences were determined by the Sanger method; and the amino acid sequences were presumed based on their base sequences, respectively.

Hereafter, this invention is further specifically described below according to examples.

Drawings referred to in Examples are briefly described as follows.

Fig. 1 is a drawing showing isotypes of monoclonal antibodies Idio 3, Idio 17, Idio 20, Idio 27 and Idio 33.

Fig. 2 is a drawing showing the monoclonal antibodies Idio 3, Idio 17, Idio 20, Idio 27 and Idio 33 specifically bind to CLN-IgG, and do not bind to other human IgGs.

Fig. 3 is a drawing showing that monoclonal antibodies Idio 3, Idio 20, Idio 27 and Idio 33 are competitively inhibiting the binding between CLN-IgG and human matrical carcinoma cell ME-180.

Fig. 4 is a drawing where the amino acid sequences of the H chain variable regions of monoclonal antibodies Idio 3, Idio 17, Idio 20, Idio 27 and Idio 33 are notated in parallel according to the Kabat's notation, and the regions of the hypervariable regions CDR1, CDR2 and CDR3 are determined.

Fig. 5 is a drawing where the amino acid sequences of the L chain variable regions of monoclonal antibodies Idio 3, Idio 17, Idio 20, Idio 27 and Idio 33 are notated in parallel according to the Kabat's notation, and the regions of the hypervariable regions CDR1, CDR2 and CDR3 are determined.

Example 1: Preparation of mouse hybridomas

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100 µl of 1 mg/ml human lgG (produced by Cappel) is intraperitoneally injected to a Balb/c mouse on the first day after its birth to prepare a mouse having immunological tolerance to human lgG. Six weeks later, the mouse is immunized as follows with CLN-lgG as an antigen.

CLN-lgG purified from a culture medium of a human/human hybridoma CLN/SUZ H11 (ATCC No. HB8307) according to an ammonium sulfate precipitation method and protein A-affinity chromatography was adjusted to a concentration of 2 μ g/ μ l with physiological saline; an equal amount of complete Freund's adjuvant solution was added; and after mixing and emulsification, 100 μ l of the emulsion (corresponding to 100 μ g of CLN-lgG) was subcutaneously injected into th immunologically tolerated mouse. Thereafter, similar immunization was repeated 4 to 5 times, the murine spleen was enucleated 4 days after the final immunization and made to be spleen cells, and they were used for the following cell fusion.

A mouse parent cells NS-1 (ATCC TIB 18) and the spleen cells are washed with portions of RPMI 1640 medium not containing serum, respectively, and the both of the cells are mixed and centrifuged. 1 ml of 50% polyethylene glycol (average molecular weight.: 4,000) is added dropwise to the resultant precipitate over a period of 1 minute. 10 ml of RPMI 1640 medium is further added over a period of 3 minutes, the mixture is centrifuged at 400 x g for 5 minutes, th precipitate is suspended in 10 ml of RPMI 1640 medium containing 20% fetal bovine serum, and the suspension is spread into a 96-well microplate.

Thereafter, the cells were cultured in HAT medium for 14 to 21 days, transferred to HT medium, and finally cultured in RPMI 1640 medium containing 10% fetal bovine serum.

The antibody titers in the culture supernatants on the wells where proliferation was observed were assayed by an enzyme-labeled antibody technique; hybridoma clones secreting monoclonal antibodies which bind to CLN-IgG but not to human IgG were obtained from the appropriate wells by the limiting dilution method; and these hybridoma clones were named No. 3, No. 17, No. 20, No. 27 and No. 33.

Example 2: Determination of isotypes of the mouse antibodies

Isotypes of the antibodies secreted from the 5 mouse hybridomas obtained in Example 1 were determined as follows using a mouse monoclonal antibody isotyping kit (produced by Amersham Co.).

The mouse hybridomas are started to be cultured at a concentration each of 5 x 104/ml in portions of RPMI 1640 medium containing 10% fetal bovine serum, respectively, and 5 days later the culture supernatants are obtained, one stick portions of the typing sticks are placed in test tubes, respectively; 3 ml portions of the culture supernatants 5-fold diluted with TBS-T (Tris-buffered saline (TBS, pH 7.6) containing 0.1% Tween 20) are added thereto respectively; and

the mixtures are incubated at room temperature for 15 minutes. The culture supernatants are discarded, 5 ml portions of TBS-T are added, and the typing sticks are washed at room temperature for 5 minutes. TBS-T was discarded, and the washing was repeated once more. 3 ml portions of a p roxidase-labeled anti-mouse antibody 500-fold diluted with TBS-T are added, and the mixtures are incubated at room temperature for 15 minutes. The typing sticks are washed twice in the same manner as above; 3 ml portions of an enzyme substrate solution (obtained by adding ne drop of 30% aqueous hydrogen peroxide to 50 ml of a TBS solution of 4-chloro-1-naphtol) are added; the mixtures are subjected to reaction at room temperature for 15 minutes; and then the sticks are washed with distilled water. The isotypes of th mouse antibodies are determined based on the resultant signals, respectively.

As a result, as shown in Fig. 1, all the isotypes of these antibodies were $\gamma 1$ and κ .

Example 3: Examination of specificities of the anti-idiotypic antibodies

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It was examined according to a dot blot technique, using an ECL Western blotting detecting reagent (produced by Amersham Co.), that the mouse anti-CLN-IgG idiotype antibodies specifically bind to CLN-IgG. The process is stated below.

CLN-IgG and human IgG1 (produced by Protogen Co.) were diluted with PBS to concentrations of 50 to 0.2 µl/ml, respectively. 2 µl portions of the thus prepared samples were spotted on a number of Hybond-ECL nitrocellulose membrane (produced by Amersham Co.), respectively and after being dried, the nitrocellulose membranes were allowed to stand at room temperature for one hour in PBS-T (0.3% Tween 20-containing PBS) containing 5% skim milk. After being washed with PBS-T, the nitrocellulose membranes were allowed to stand at room temperature for one hour in the culture supernatants (500-fold diluted with PBS-T) of mouse hybridomas No. 3, No. 17, No. 20, No. 27 and No. 33, respectively. After being washed with PBS-T, the nitrocellulose membranes were allowed to stand at room temperature for one hour in portions of a peroxidase-labeled sheep anti-mouse Ig antibody 3,000-fold diluted with PBS-T, respectively. After being washed with PBS-T, the nitrocellulose membranes were subjected to reaction for one minute in portions of the ECL detecting reagent, and sheets of X-ray film were exposed for 30 seconds to the light emitted from the resultant nitrocellulose membranes, respectively.

The results of the sheets of X-ray film developed are shown in Fig. 2. Any of the five antibodies bound to CLN-IgG, but did not bind to human IgG1. Namely, it was revealed that these antibodies are specific to CLN-IgG.

Next, it was examined whether or not the mouse antibodies have an activity to inhibit the binding of a human monoclonal antibody CLN-IgG to a human cancer cell. The method is stated below.

A human cervical carcinoma cell ME-180 (available from ATCC) is cultured in DF medium (a 1:1 mixed medium of DME: F-12) containing 10% fetal bovine serum. At the stage when the number of the cells becomes 5 x 106 to 1 x 107, the cells are detached from the bottom face of the Petri dish using trypsin, collected by centrifugation and sufficiently washed with the medium. A constant number (105/100 µl) each of the cells is placed in each well of a 96-well microtiter plate, and allowed to stand at 37°C overnight to be attached on the plate. 50 µl portions of 3% glutaraldehyde solution were added dropwise into the respective wells, and the mixtures are allowed to stand at 37°C for 20 minutes to fix the cells. The cells of each well are centrifuged at 200 x g for 10 minutes and washed three times with a gelatin buffer (10 mM phosphate-buffered physiological saline containing 0.3% gelatin); 200 μl portions of 1% bovine serum albumin (BSA) solution are added dropwise; and the mixture is allowed to stand at 37°C for one hour to block the plate. The cells are washed three times with the gelatin buffer to remove BSA not adsorbed. Thereafter, dilutions at various rates (100 to 1,000,000-fold) of the ascites obtained by intraperitoneally inoculating into mice the various hybridomas secreting the mouse anti-idiotypic antibodies are added dropwise together with CLN-IgG (50 µg each), and the mixtures are subjected to reaction at 37°C for one hour. The cells of these wells are washed three times with the gelatin buffer, 50 µl portions of a 3,000-fold diluted peroxidase-conjugated goat anti-human Ig antibody (produced by TACO Co.) are added dropwise, respectively, and the mixtures are subjected to reaction at 37°C for 30 minutes. The cells are washed three times with the gelatin buffer, and portions of a substrate solution containing hydrogen peroxide and o-phenylenediamine are added to perform reaction in a darkroom. 10 minutes later, 50 µl portions of 5N sulfuric acid are added to stop the reaction. When the peroxidase-conjugated goat anti-Ig antibody remains on the microplate, namely when the human IgG to be bound thereto remains, a yellow reaction product having absorption at 490 nm is formed. The amount of CLN-IgG bound to the cancer cell is determined by measuring the amount of the reaction product by a spectrometer.

It was clarified, according to the above method, that all the mouse antibodies Idio 3, Idio 17, Idio 20, Idio 27 and Idio 33 inhibit the binding of CLN-IgG to the cancer cell (Fig. 3).

From the foregoing, these mouse antibodies are antibodies against the idiotypes of CLN-IgG.

55 Example 4: Preparation of RNA

From the five kinds of mouse hybridomas No. 3, No. 17, No. 20, No. 27 and No. 33, the cytoplasmic RNAs were extracted according to the method disclosed in Molecular Cloning (2nd edition, edited by Sambrook et al., Cold Spring Harbor Laboratory Press 1989) 7, 12, as stated below.

108 each of the hybridomas cells are collected by centrifugation, and washed twice with 10 times each precipitate's volume of a phosphate-buffered saline. The cills of these groups are centrifuged at 2,000 x g and 4°C for 5 minutes, and the resultant precipitates are suspended in 200 µl portions of an RNA extracting solution (0.14 M NaCl, 1.5 mM MgCl₂, 10 mM Tris-HCl pH 8.6, 0.5% Nonidet P-40, 1 mM dithiothreitol, 20 mM vanadylribonucleoside complex), respectively. The susp nsions are subjected to vortex for 15 seconds and allowed to stand on ice for 5 minutes. The resultant suspensions are centrifuged at 12,000 x g for 30 seconds to remove the cill nuclei as precipitates; to the supernatants are, respectively, added 200 μl portions of a proteinase buffer (0.2 M Tris-HCl pH 8.0, 25 mM EDTA pH 8.0, 0.3 M NaCl, 1.2% SDS) and 1 µl portions of an aqueous proteinase K solution (20 mg/ml); and the mixtures are sufficiently stirred and subjected to incubation at 37°C for 30 minutes. Equal volume portions of phenol/chloroform are added to the reaction solutions, respectively, and the mixtures are stirred, centrifuged at 5,000 x g and room temperature for 10 minutes, and then allowed to separate into organic layers and aqueous layers, respectively. 400 µl portions of isopropanol cooled on ice in advance are added to the aqueous layers recovered, respectively, and the mixtures are allowed to stand on ice for 30 minutes. The mixtures are centrifuged at 12,000 x g and 4°C for 10 minutes to collect RNAs. The resultant RNA precipitates are washed with 1 ml portions of ethanol, dried under reduced pressure and suspended in appropriate amount portions of TE buffer, respectively. Using the cytoplasmic RNAs obtained according to the above operations, the antibody genes are amplified.

Example 5: Amplification and cloning of the antibody genes by the RT-PCR method

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The antibody genes were amplified from the cytoplasmic RNAs obtained in Example 4, using a GeneAmp® RNA PCR kit (produced by Takara Shuzo Co., Ltd.). First, 20 μl each of reactive solutions were prepared containing PCR buffer II (x1), 5 mM MgCl₂, 1 mM dATP, 1 mM dGTP, 1 mM dTTP and 1 mM dCTP, 1 U/μl an RNase inhibitor, 2.5 μM a random hexamer, 2.5 U/μl a reverse transcriptase and 100 ng each of the above-mentioned cytoplasmic RNAs, respectively; 20 μl portions of a mineral oil were overlaid thereon respectively; and incubations were performed at room temperature for 10 minutes, at 42°C for 15 minutes, at 99°C for 5 minutes and then at 4°C for 5 minutes to perform cDNA synthesis by reverse transcription reaction. Then, 80 μl portions of a solution consisting of 4 μl of 25 mM MgCl₂, 8 μl of 10x PCR buffer II, 65.5 μl of sterile distilled water, 0.5 μl of AmpliTaq DNA polymerase (5 U/μl) and 2 μl of PCR primers (each 100 pmoles) were added to the above 20 μl of the reverse transcription reaction solutions; 80 μl portions of the mineral oil were overlaid thereon; and PCR reactions were succeedingly performed. Each reaction was performed by repeating 30 times the cycle of 94°C for 1.5 minutes, 50°C for 2 minutes and then 72°C for 3 minutes. The base sequences of the PCR primers are shown below. The primers contained in a lg-Prime™ kit (produced by Novagen Co.) were used except for the primer of the leader sequence C for H chains.

Primer for H chains	
Leader sequence A	5' GGGAATTCATGRASTTSKGGYTMARCTKGRTTT 3'
Leader sequence B	5' GGGAATTCATGRAATGSASCTGGGTYWTYCTCTT 3'
Leader sequence C	5' TTAAATGGTATCCAGTGT 3'
Constant region	5' CCCAAGCTTCCAGGGRCCARKGGATARACIGRTGG 3'

Primer for L chains	
Leader sequence A	5' GGGAATTCATGRAGWCACAKWCYCAGGTCTTT 3'
Leader sequence B	5' GGGAATTCATGGAGACAGACACACTCCTGCTAT 3'
Constant region	5' CCCAAGCTTACTGGATGGTGGGAAGATGGA 3'

In the above, the alphabets other than A, G, C and T mean the following bases. R=A/G, W=A/T, I=inosin , Y=C/T, D=A/G/T, K=G/T, H=A/C/T, S=C/G, V=A/C/G, M=A/C, B=G/C/T

10 µl portions of the resultant 100 µl each of the PCR reaction products are subjected to 1.5% agarose gel electrophoresis, and it was confirmed that the antibody general fragments each about 600 bp long were amplified. As a result, in the case of the H chains, the antibody genes derived from No. 3 and No. 17 were amplified in the leader sequence A, the antibody genes derived from No. 20 and No. 27 were amplified in the leader sequence B, and the antibody gene derived from No. 33 was amplified in the leader sequence C. On the other hand, in the L chains, the antibody genes derived from No. 27 and No. 33 were amplified in the case where the leader sequence A was used, and the antibody genes derived from No. 3, No. 17 and No. 20 were amplified in the leader sequence B.

Each of the PCR-amplified fragments about 600 bp long was integrated into pCR 1000 vector or pCR^{IM} vector using TA cloning kit (produced by Invitogen Co.). Specifically, ligation mix solutions were prepared by mixing 1 μ l portions of the PCR reaction products, 1 μ l portions of 10 x the ligation buffer, 2 μ l portions of pCR1000 or pCR $^{\mathrm{IM}}$ vector (corresponding to 50 μ g), 1 μ l of T4 DNA ligase and 6 μ l portions of sterilized water, respectively; and incubated overnight at 12°C. Separately, 50 μ l portions of a suspension of a competent Escherichia coli INV α T strain, to which portions were added 2 μ l portions of 0.5 M β -mercaptoethanol, respectively, were prepared; and 1 μ l portions of the above ligation mix solutions are added thereto, respectively. The mixtures are allowed to stand on ice for 30 minutes, incubated at 42°C for one minute, and rapidly cooled on ice for 2 minutes. 450 μ l portions of SOC medium warmed to 42°C in advance were added to the resultant Escherichia coli solutions, respectively, and the mixtures are cultured with shaking at 37°C for one hour. Meanwhile, 25 μ l portions of X-Gal (40 mg/ μ l) are spreaded onto a number of LB agar plates each containing Kanamycin (50 μ g/ μ l), respectively, and the agar plates are incubated at 37°C until each X-Gal completely permeates the agar plate.

200 µl portions of the <u>Escherichia coli</u> culture broths after completion of culture were spread on the agar plate dried, respectively, and the plates were allowed to stand at 37°C overnight to give white colonies each having Kanamycin resistance.

Plasmids were purified from the <u>Escherichia coli</u> clones containing the respective antibody genes, and named 3KB11, 17KB1, 20KB1, 27KA2, 33KA26, 3GB1, 17GB7, 20GA2, 27GA5 and 33GC003, respectively. Purification of the plasmids is performed as follows.

The Escherichia coli strains containing the above plasmids, respectively, are cultured 37°C overnight in 100 ml portions of LB medium containing Kanamycin (50 µg/ml), respectively. Each of the resultant culture broths is centrifuged at 3,000 rpm for 10 minutes; the cells collected are suspended in 3 ml of an ice-cooled suspension (50 mM glucose, 10 mM EDTA, 2 mM Tris-HCl pH 8.0); and the suspension is allowed to stand at room temperature for 5 minutes. 6 ml of an alkali lysing solution (0.2 N sodium hydroxide, 1% SDS) is added, and the mixture is mixed by gently turning the centrifugation vessel upside down, and allowed to stand on ice for 5 minutes. 4.5 ml of an ice-cooled neutralizing solution (5 M potassium acetate pH 4.8) is added, and the mixture is centrifuged at 12,000 rpm and 4°C for 10 minutes. The supernatant is transferred into another centrifugation vessel; 1 ml of heat-treated 100 µg/ml RNase A solution is added; and the mixture is subjected to reaction for one hour in an incubator of 37°C to perform RNA digestion. To the reaction solution are added 6 ml of TE buffer-saturated phenol and 6 ml of chloroform/isoamyl alcohol (24:1), and the mixture is subjected to vortex for 30 seconds and then centrifuged at 10,000 rpm and 4°C for 3 minutes. The aqueous layer is transferred into another centrifugation vessel, an equal amount of isopropanol is added, and the mixture is sufficiently mixed and then centrifuged at 10,000 rpm and room temperature for 10 minutes.

The resultant precipitate is washed with 1 ml of 70% cold (-20°C) ethanol, dried under reduced pressure, and dissolved in 480 μ l of sterilized water. The solution is transferred into an Eppendorf tube; 120 μ l of 4 M NaCl and 600 μ l of 13% polyethylene glycol #6000 are added; and the mixture is allowed to stand on ice for 20 minutes. The mixture is then centrifuged at 10,000 rpm and 4°C for 10 minutes, and the precipitate is washed with 1 ml of 70% cold (-20°C) ethanol, dried under reduced pressure and dissolved in 100 μ l of TE buffer. The resultant purified plasmid was used as a template for sequencing reaction.

Example 6: Determination of the base sequences

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Sanger reactions were performed using as templates the plasmids cloning purified in Example 5 and a fluorescencelabeled primer; the reaction products were analyzed by a DNA sequencer DSQ-1 (produced by Shimadzu Corporation); and the DNA base sequences of the insert parts of the plasmids were also determined.

The sequencing reactions were performed using AmpliTaq cycle sequencing kit (produced by Takara Shuzo Co., Ltd.) and a fluorescence-labeled primer in a reagent kit (produced by Wakunaga Pharmaceutical Co., Ltd.) exclusively used for a fluorencene-type DNA sequencer. First, 2 to 4 µg of one of the plasmids purified as stated in Exampl 5 is mixed with 1 µl of the FITC-labeled primer (1 p mole/µl, forward or reverse is used) and 2 µl of the 10 x cycling mix solution, and sterilized water is added to prepare 10 µl in final volume of a reaction mix. Four tubes are prepared in which 2 µl portions of the termination mix (A, G, C, T) were placed in advance, respectively. 2 µl portions of the above reaction mix were taken and placed into the respective tubes. The mixtures are corrected by centrifugation, 10 µl portions of a mineral oil are overlaid, and cycling reactions are performed under the following conditions; Precycle 95°C, 3 minutes; first cycle 95°C 30 seconds, 60°C 30 seconds, 72°C 1 minute (repeated 15 times); second cycle 95°C 30 seconds, 72°C 1 minute (repeated 15 times); postcycle 4°C.

2 μl portions of a reaction-stopping dye solution (95% formaldehyde, 20 mM EDTA, 0.05% methyl violet) are added, and the mixtures are mixed by centrifugation and preserved at 20°C until they are electrophoresed.

As 5% polyacrylamide gel was used one obtained by adding pure water to $50\,\mathrm{g}$ of urea, 6 ml of 10 x TBE buff r (0.89 M Tris-HCl, 0.89 M boric acid, 0.025 M EDTA disodium salt) and 10 ml of 30% acrylamide solution (28.5% acrylamide and 1.5% methylenebisacrylamide, both produced by BIO-RAD Co.) to make the wholevolume 60 ml; filtering the mixture with 0.22- μ m filter; deaerating the filtrate for 30 minutes; adding 150 μ l of 10% ammonium persulfat and 15 μ l of TEMEO; allowing the mixture to stand overnight to make it g. l.

The gel was set in the DNA sequencer DSQ-1, and prerun was performed at a constant voltage of 1,000 V for on hour. Each of the samples was dinatured at 95°C for 3 minutes immediately before electrophoresis, and rapidly cooled on ice, and 2 to 3 µl of the reaction solution was sucked up from this bottom part of the tube by a micro-syringe and loaded onto the gel. Samples run was performed at a constant electric power of 20 W for 12 hours.

After completion of electrophoresis, the base sequence was determined using the software attached to DSO-1. The sequence was confirmed by sequencing both of the sense and antisense chains of the same plasmid from both directions.

The resultant base sequences of the variable regions of the H chains and L chains of the five kinds of the mouse monoclonal antibodies, and amino acid sequences presumed therefrom are shown in the following sequence listing. Relation between the sequence numbers and the sequences of the clones are as follows:

15 Sequence No. 1: Idio 3 H chain variable region (clone 3GB1)

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Sequence No. 2: Idio 17 H chain variable region (clone 17GB7)

Sequence No. 3: Idio 20 H chain variable region (clone 20GA2)

Sequence No. 4: Idio 27 H chain variable region (clone 27GA5)

Sequence No. 5: Idio 33 H chain variable region (clone 33GC003)

Sequence No. 6: Idio 3 L chain variable region (clone 3KB11)

Sequence No. 7: Idio 17 L chain variable region (clone 17KB1)

Sequence No. 8 : Idio 20 L chain variable region (clone 20KB1)

Sequence No. 9: Idio 27 L chain variable region (clone 27KA2)

Sequence No.10: Idio 33 L chain variable region (clone 33KA26)

Example 7 Determination of hypervariable regions

The amino acid sequences obtained in Example 6 were notated in parallel according to the numbering of Kabat et al.'s data base (Sequences of proteins of immunological interest Fifth edition, U. S. Department of health and human services. Public health service, National Institutes of Health. NIH Publication No. 91-3242, Kabat et al. 1991), and the amino acid sequences of the hypervariable regions CDR1, CDR2 and CDR3 of each antibody were identified (Fig. 4, H chains, Fig. 5 L chains). In order to confirm the novelty of the identified amino acid sequences of the hypervariable regions CDR1, CDR2 and CDR3, retrieval by a computer was performed using the above Kabat et al.'s data base and a protein data base NBRF-PDB (National Biomedical Research Foundation - protein data base) Release 36.

As a result, the amino acid sequences of Idio 3 H chain CDR1, Idio 17 H chain CDR1, Idio 20 H chain CDR1, Idio 27 H chain CDR1, Idio 33 H chain CDR2, Idio 3 L chain CDR2, Idio 17 L chain CDR2, Idio 27 L chain CDR2 and Idio 33 L chain CDR2 were the same as those of known antibodies, but the amino acid sequences of other CDRs were

revealed to be novel sequences.

	Sequence Listing	
5	Seq. I.D. number : 1	
	Sequence length: 399	
	Sequence type : nucleic acid	
	Strandedness : double	
10	Topology : linear	
	Sequence kind: mRNA	
	Original source	
	Organism : mouse	
15	Sequence characteristics	
	Symbol expressing characteristics : CDS	
	Presence position: 1399	
	Characteristics determination method: S	
20		
	Symbol expressing characteristics : sig peptide	
	Presence position: 127	
	Characteristics determination method : S	
25	Sequence	
	CTG TCG GTA ACT TCA GGG GTC TAC TCA GAG GTT CAG CTC CAG CAG TCT Leu Ser Val Thr Ser Gly Val Tyr Ser Glu Val Gln Leu Gln Gln Ser	48
	-5 · 1 5	
	GGG ACT GTG CTG GCA AGG CCT GGG GCT TCA GTG AAG ATG TCC TGC AAG	96
30	Gly Thr Val Leu Ala Arg Pro Gly Ala Ser Val Lys Met Ser Cys Lys	
	10 15 20 GCT TCG GGC TAC ACC TTT AAC AGC TAC TGG ATG CAC TGG GTA AAA CAG	144
•	Ala Ser Gly Tyr Thr Phe Asn Ser Tyr Trp Met His Trp Val Lys Gln	144
	25 30 35	
35	AGG CCT GGA CAG GGT CTG GAA TGG ATT GGC GCG ATT TAT CCT GGA AAT	192
	Arg Pro Gly Gln Gly Leu Glu Trp Ile Gly Ala Ile Tyr Pro Gly Asn	
	40 45 50 55 AGT GAT ATT AGC TAC AGC CAG AAC TIT AAG GAC AGG GCC AAA CTG ACT	240
40	Ser Asp Ile Ser Tyr Ser Gln Asn Phe Lys Asp Arg Ala Lys Leu Thr	
40	60 65 70	
	GCC GTC ACA TCC ACC AGC ACT GCC TAC ATG GAA CTC AGA AGC CTG ACA	288
	Ala Val Thr Ser Thr Ser Thr Ala Tyr Met Glu Leu Arg Ser Leu Thr 75 80 85	
45	AAT GAG GAC TOT GOG GTO TAT TTO TGT ACA AAA GAG GAA TAT GAT TAC	336
	Asn Glu Asp Ser Ala Val Tyr Phe Cys Thr Lys Glu Glu Tyr Asp Tyr	
	90 . 95 100	
	GAC ACC CTG GAC TAC TGG GGT CAA GGA ACC TCA GTC ACC GTC TCC TCA	384
50	Asp Thr Leu Asp Tyr Trp Gly Gln Gly Thr Ser Val Thr Val Ser Ser	
	GCC AAA ACG ACA CCC	399
	Ala Lys Thr Thr Pro	
	120	

	Sequence Listing	
<i>5</i>	Seq. I.D. number : 2	
J	Sequence length: 402	
	Sequence type : nucleic acid	
	Strandedness : double	
10	Topology : linear	
	Sequence kind: mRNA	
	Original source	
	Organism : mouse	
15	Sequence characteristics	
	Symbol expressing characteristics: CDS	
	Presence position: 1402	
20	Characteristics determination method: S	
•	Symbol expressing characteristics : sig peptide	
	Presence position : 130	
	Characteristics determination method : S	
25	Sequence	
	ATT CTG TCG GTA ACT TCA GGG GTC TAC TCA GAG GTT CAG CTC CAG CAG	48
	Ile Leu Ser Val Thr Ser Gly Val Tyr Ser Glu Val Gln Leu Gln Gln -10 -5 1	
	TCT GGG ACT GTG CTG GCA AGG CCT GGG GCT TCA GTG AAG ATG TCC TGC	96
3 <i>0</i> .	Ser Gly Thr Val Leu Ala Arg Pro Gly Ala Ser Val Lys Met Ser Cys	36
	10 15 20	
	AAG GCT TCG GGC TAC ACC TTT AAC AGC TAC TGG ATG CAC TGG GTA AAA	14
	Lys Ala Ser Gly Tyr Thr Phe Asn Ser Tyr Trp Met His Trp Val Lys 25 30 35	
35	CAG AGG CCT GGA CAG GGT CTG GAA TGG ATT GGC GCG ATT TAT CCT GGA	192
	Gln Arg Pro Gly Gln Gly Leu Glu Trp Ile Gly Ala Ile Tyr Pro Gly	194
	40 45 50	
	AAT AGT GAT ATT AGC TAC AGC CAG AAC TTT AAG GAC AGG GCC AAA CTG	240
0	Asn Ser Asp Ile Ser Tyr Ser Gln Asn Phe Lys Asp Arg Ala Lys Leu 55 60 65	
	ACT GCC GTC ACA TCC ACC AGC ACT GCC TAC ATG GAA CTC AGA AGC CTG	288
	Thr Ala Val Thr Ser Thr Ser Thr Ala Tyr Met Glu Leu Arg Ser Leu	200
	70 75 80 85	
5	ACA AAT GAG GAC TCT GCG GTC TAT TTC TGT ACA AAA GAG GAA TAT GAT	336
	Thr Asn Glu Asp Ser Ala Val Tyr Phe Cys Thr Lys Glu Glu Tyr Asp	
	TAC GAC ACC CTG GAC TAC TGG GGT CAA GGA ACC TCA GTC ACC GTC TCC	384
	Tyr Asp Thr Leu Asp Tyr Trp Gly Gln Gly Thr Ser Val Thr Val Ser	304
0	105 110 115	
	TCA GCC AAA ACG ACA CCC	402
	Ser Ala Lys Thr Thr Pro	

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.5	Seq.	1.	D. r	umb	er	: 3											
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	Sequ				010.	,											
	-			GGG	CTA	AAC	TGG	GTT	TTC	CTT	GTA	ACA	CIT	TTA	AAT	GGT	48
25	Met	Glu	Phe	Gly	Leu	Asn	Trp	Val	Phe	Leu	Val	Thr	Leu	Leu	Asn	Gly	
					-15					-10					-5		
																CAG Gln	. 96
	116	GIII	Cys	1	VAI	rys	Den	Val	5	Ser	GLY	GIY	GLY	10		GIII	
30	CCT	GGG	GGT	TCT	CTC	AGA	CTC	TCC	TGT	GCA	ACT	TCŤ	GGG	TTA	ACC	TTC	144
	Pro	Gly	Gly	Ser	Leu	Àrg	Leu	Ser	Суѕ	Ala	Thr	Ser	Gly	Leu	Thr	Phe	
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35		30	-,-	-,,-			35		3			40	•	-,-			
	GAA	TGG	TIG	GGT	TTT	ATT	AGA	AAC	AAA	GCT	AAT	CTT	TAC	ACA	ACA	GAC	240
	Glu	Trp	Leu	Gly	Phe		Arg	Asn	Lys	Ala		Leu	Tyr	Thr	Thr	-	
40	45 TAC	АСТ	GCA	יור־אני	GTG	50 AAG	CCT	ന്ദ്ര	Tire	ACC	55 ATC	TCC	AGA	GAT	AAT	60 CCC	288
40	Tyr										_						
					65		_			70					75		
	CAA																336
40	Gln	Ser		Leu 80	Tyr	Leu	Gln	Met	Asn 85	Thr	Leu	Thr	Thr	Glu 90	Asp	Ser	
45	GCC				TGT	GCA	AGA	GAT		GGG	GGG	AGG	GAC	_	TAC	TTC	384
	Ala										_						
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	Sequence Listing	
_	Seq. I.D. number : 4	
5	Sequence length: 411	
	Sequence type : nucleic acid	
	Strandedn ss : double	
**	Topology : linear	
10	Sequence kind: mRNA	
•	Original source	
	Organism : mouse	
	Sequence characteristics	
15	Symbol expressing characteristics : CDS	
	Presence position: 1411	
	Characteristics determination method : S	
20	Symbol expressing characteristics: sig peptide	
	Presence position: 130	
	Characteristics determination method : S	
	Sequence	
25	CTT GTA ACA CGT TTA AAT GGT ATC CAG TGT GAG GTG AAG CTG GTG GAG	48
	Leu Val Thr Arg Leu Asn Gly Ile Gln Cys Glu Val Lys Leu Val Glu	
	TCT GGA GGC GGT GGA CAG CCT GGG GGT TCT CTG AGA CTC TCC TGT	96
	Ser Gly Gly Leu Val Gln Pro Gly Gly Ser Leu Arg Leu Ser Cys	90
30	10 15 20	
	GCA ACT TCT GGG TTC ACC TTC ACT GAT TAC TAC ATG AAC TGG GTC CGC	144
	Ala Thr Ser Gly Phe Thr Phe Thr Asp Tyr Tyr Met Asn Trp Val Arg	
	CAG CCT CCA GGA AAG GCA CTT GAG TGG TTG GGT TTT ATT AGA AAC AAA	192
35	Gln Pro Pro Gly Lys Ala Leu Glu Trp Leu Gly Phe Ile Arg Asn Lys	
	40 45 50	
	GCT AAT TAT TAC ACA ACA GAG TAC AGT GCA TCT GTG AAG GGT CGG TTC	240
	Ala Asn Tyr Tyr Thr Thr Glu Tyr Ser Ala Ser Val Lys Gly Arg Phe 55 60 65	
ю	ACC ATC TCC AGA GAT AAT TCC CAA AGC ATC CTC TAT CTT CAA ATG AAC	288
	Thr Ile Ser Arg Asp Asn Ser Gln Ser Ile Leu Tyr Leu Gln Met Asn	÷00
	70 75 80 85	
	ACC CTG AGA GCT GAG GAC AGT GCC ACT TAT TAC TGT GCA AGA GAT GGG	336
15	Thr Leu Arg Ala Glu Asp Ser Ala Thr Tyr Tyr Cys Ala Arg Asp Gly 90 95 100	
	TTC CTA CGG GAC TGG TAC TTC GAT GTC TGG GGC GCA GGG ACC ACG GTC	384
	Phe Leu Arg Asp Trp Tyr Phe Asp Val Trp Gly Ala Gly Thr Thr Val	-
	. 105 110 115	
io	ACC GTC TCC TCA GCC AAA ACG ACA CCC	411
	Thr Val Ser Ser Ala Lys Thr Thr Pro	411
	120 125	

	Sequence 1	Listing					
5	Seq. I.D.	number :	5				
	Sequence]	length:	363				
	Sequence t	type : nu	cleic aci	d			
	Strandedne						
10	Topology:						
	Sequence k		NΔ				
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15		sm : mous	Δ.				
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		·	ng charac		es : CDS		
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	Sequence				-		
25					CTG GCA AGA		
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					Tyr Thr Phe		
30		20		25	•	30	
					CAG GGT CTG		
	Trp Met Glr		Lys Gln Arg		31n Gly Leu	_	e
35	ככב כריי איזיי	35 יידאייר רכייירים	ድርስ ርስጥ ር ርጥ	40 Cam acm 2	AGG TAC ACT	45	2
					arg Tyr Thr		
	50		55		60		-
	AAG GGC AAG	GCC ACA T	TG ACT GCA	GCT AAA T	OOA TOC AGC	ACA GCC TAG	240
40		Ala Thr L		Ala Lys S	Ser Ser Ser	Thr Ala Tyr	•
	65 ATC CAA CTC	LACC ACC T	70	CNC CNC T	75	Mam mag mon	
	ATG CAA CTC						
45	80		5		0	95 - 171 - 171 95	•
	GCA AGA TCG	GGC TAC T	AT GGT AGC	TTC GTT G	GG TTT GCT	TAC TGG GGC	336
	Ala Arg Ser	Gly Tyr T	yr Gly Ser	Phe Val G	ly Phe Ala	Tyr Trp Gly	
	O	100		105	,	110	
<i>50</i>	CAA GGG ACT						. 363
	Gln Gly Thr	Leu vai T	iir val Ser	120			
		~=~		120	•		

	•											
	Sequence Listing											
5	Seq. I.D. number : 6											
	Sequence length: 354											
	Sequence type : nucleic acid											
	Strandedness : double											
10	Topology : linear	•										
	Sequence kind: mRNA											
	Original source											
15	Organism : mouse											
,,,												
	Sequence characteristics											
	Symbol expressing characteristics : CDS											
20	Presence position: 1354											
	Characteristics determination method : S											
	Sequence											
os.	GAC ATT GTG CTG ACA CAG TCT CCT GCT TCC TTA GCT GTA TCT CCT CTG	48										
25	Asp Ile Val Leu Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Pro Leu											
	1 5 10 15 GGG CAG AGG GCC ACC ATC TCA TAC AGG GCC AGC AAA AGT GTG CAG TTA											
	Gly Gln Arg Ala Thr Ile Ser Tyr Arg Ala Ser Lys Ser Val Gln Leu	96										
30	20 25 30											
	CAT CTG GCT ATA GTT TAT ATG CAC TGG AAC CAA CAG AAA CCA GGA CAG	144										
	His Leu Ala Ile Val Tyr Met His Trp Asn Gln Gln Lys Pro Gly Gln											
	35 40 45 CCA CCC AGA CTC CTC ATC TAT CTT GTA TCC AAC CTA GAA TCT GGG GTC											
35	Pro Pro Arg Leu Leu Ile Tyr Leu Val Ser Asn Leu Glu Ser Gly Val	192										
	50 55 60											
	CCT GCC AGG TTC AGT GGC AGT GGG TCT GGG ACA GAC TTC ACC CTC AAC	240										
40	Pro Ala Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Asn											
	65 70 75 ATC CAT CCT GTG GAG GAG GAG GAT GCT GCA ACC TAT TAC TGT CAG CAC											
	Ile His Pro Val Glu Glu Glu Asp Ala Ala Thr Tyr Tyr Cys Gln His	288										
	80 85 90 95											
45	ATT AGG GTA GCT TAC ACG TTC GGA GGG GGG ACC AAG CTG GAA ATA AAA	336										
	Ile Arg Val Ala Tyr Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys											
,	100 105 110 CGG GCT GAT GCT GCA CCA											
50	Arg Ala Asp Ala Ala Pro	354										
	115											

	bedreite Bioling	
5	Seq. I.D. number : 7	
	Sequence length: 438	
	Sequence type : nucl ic acid	
	Strandedness : double	
	Topology : linear	
10	Sequence kind: mRNA	
	Original source	
	•	
	Organism : mouse	
15	Sequence characteristics	
	Symbol expressing characteristics : CDS	
	Presence position: 1438	
	Characteristics determination method : S	
20	Symbol expressing characteristics : sig peptide	
	Presence position: 139	
	Characteristics determination method : S	
•	Sequence	
25	CTA TGG GTA CTG CTC TGG GTT CCA GGT TCC ACT GGT GAC ATT GTG	48
	Leu Trp Val Leu Leu Trp Val Pro Gly Ser Thr Gly Asp Ile Val	
	-10 -5. 1	
	CTG ACA CAG TCT CCT GCT TCC TTA GCT GTA TCT CTG GGG CAG AGG GCC	96
30	Leu Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Leu Gly Gln Arg Ala 5 10 15	
	TCC ATC TCA TAC AGG GCC AGC AAA AGT GTC AGT ACA TCT GGC TAT AGT	144
	Ser Ile Ser Tyr Arg Ala Ser Lys Ser Val Ser Thr Ser Gly Tyr Ser	
	20 25 30	
35	TAT ATG CAC TGG AAC CAA CAG AAA CCA GGA CAG CCA CCC AGA CTC CTC	192
33	Tyr Met His Trp Asn Gln Gln Lys Pro Gly Gln Pro Pro Arg Leu Leu 35 40 45 50	
	ATC TAT CIT GTA TCC AAC CTA GAA TCT GGG GTC CCT GCC AGG TTC AGT	240
	Ile Tyr Leu Val Ser Asn Leu Glu Ser Gly Val Pro Ala Arg Phe Ser	240
	55 60 65	
40	GGC AGT GGG TCT GGG ACA GAC TTC ACC CTC AAC ATC CAT CCT GTG GAG	288
	Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Asn Ile His Pro Val Glu	
	70 75 80 GAG GAG GAT GCT GCA ACC TAT TAC TGT CAG CAC ATT AGG GGA GCT TAC	226
	Glu Glu Asp Ala Ala Thr Tyr Tyr Cys Gln His Ile Arg Gly Ala Tyr	336
45	85 90 95	
•	ACG TTC GGA GGG GGG ACC AAG CTG GAA ATA AAA CGG GCT GAT GCT GCA	384
•	Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys Arg Ala Asp Ala Ala	
	100 105 110	
50	CCA ACT GTA TCC ATC TTC CCA CCA TCC AGT AAG CTT GGG AAA CGG TTC Pro Thr Val Ser Ile Phe Pro Pro Ser Ser Lys Leu Gly Lys Arg Phe	432
•	115 120 125 130 125 130	
		438
	Ala Pro	

	Sequ nce Listing	
5	Seq. I.D. number : 8	
3	Sequence length: 417	
	Sequence type : nucleic acid	
	Strandedness : double	
10	Topology : linear	
	Sequence kind: mRNA	
	Original source	
	Organism : mouse	
15	Sequence characteristics	
1	Symbol expressing characteristics : CDS	
	Presence position: 28417	
	Characteristics determination method : S	
20	Symbol expressing characteristics : sig peptide	
	Presence position: 2890	
	Characteristics determination method : S	
25 .	Sequence GGCCGCG GTGAGAACCG TTGGGAATTC ATG GAG ACA GAC ACA CTC CTG	
	Met Glu Thr Asp Thr Leu Leu	48
	-20 -15	
•	CTA TGG GTA CTG CTG CTC TGG GTT CCA GGT TCC ACT GGT GAC ATT GTG	96
30	Leu Trp Val Leu Leu Trp Val Pro Gly Ser Thr Gly Asp Ile Val	
	-10 -5 1 CTG ACA CAG TCT CCT GCT TCC TTA GCT GTA TCT CTG GGG CAG AGG GCC	144
	Leu Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Leu Gly Gln Arg Ala	744
	5 10 15	
35 ·	ACC ATC TCA TAC AGG GCC AGC AAA AGT GTC AGT ACA TCT GGC TAT AGT	192
	Thr Ile Ser Tyr Arg Ala Ser Lys Ser Val Ser Thr Ser Gly Tyr Ser	
	20 25 30 TAT ATG CAC TGG AAC CAA CAG AGA CCA GGA CAG CCC AGA CTC CTC	240
	Tyr Met His Trp Asn Gln Gln Arg Pro Gly Gln Pro Pro Arg Leu Leu	
40	35 40 45 50	
	ATC TAT CTT GTA TCC AAC CTA GAC TCT GGG GTC CCT GCC AGG TTC AGT	288
•	Ile Tyr Leu Val Ser Asn Leu Asp Ser Gly Val Pro Ala Arg Phe Ser 55 60 65	
	55 60 65 GGC AGT GGG TCT GGG ACA GAC TTC ACC CTC AAC ATC CAT CCT GTG GAG	336
45	Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Asn Ile His Pro Val Glu	
	70 75 80	
	GAG GAG GAT GCT GCA ACC TAT TAC TGT CAG CAC ATT GAG GGA GCT TAC	384
	Glu Glu Asp Ala Ala Thr Tyr Tyr Cys Gln His Ile Glu Gly Ala Tyr 85. 90 95	
50	ACG TTC GGA GGG GGG ACC AAG CTG GAA ATA AAA	417
	Thr Phe Gly Gly Thr Lys Leu Glu Ile Lys	
	100 105	

	Sequ	enc	e L	isti	ng												
•	Seq.	I.	D. 1	umb	er	: 9	•										
5	Sequ	enc	e 1	engt	h :	420)										
Sequence Listing Seq. I.D. number: 9 Sequence length: 420 Sequence type: nucleic acid Strandedness: double Topology: linear Sequence kind: mRNA Original source Organism: mouse Sequence characteristics Symbol expressing characteristics: CDS Presence position: 31420 Characteristics determination method: S Symbol expressing characteristics: sig peptide Presence position: 3190 Characteristics determination method: S Sequence Sequence GCGGCGCGG TGAGAACCGT TTGGGAATTC ATG GAG ACA CAG TCC CAG Met Glu Thr Gln Ser Gln -20 -15 GTC TTT GTA TTC GTG TTT CTC TGG TTG TCT GGT GTT GAC GGA GAC ATT Val Phe Val Phe Val Phe Leu Trp Leu Ser Gly Val Asp Gly Asp Ile -10 -5 GTC ATG ACC CAG TCT CAC AAA TTC ATG TCA CTA GTA GGA GAC ACG Val Met Thr Gln Ser His Lys Phe Met Ser Thr Ser Val Gly Asp Arg 5 GTC AGT ATC ACC TGC AAG GCC AGT CAG GAT GTG AAT ACT GCT GTA GCC Val Ser Ile Thr Cys Lys Ala Ser Gln Asp Val Asn Thr Ala Val Ala 20 25 GTG GTA TAC ACC AGA TCC GA GAC CAC TCC AGA TCC CTG TTG TTG TTG TTG TTG TTG TTG TTG																	
	Stra	nde	dne:	ss.:	do	uble	;										
10	Торо	log	у:	lin	ear				•								
	Sequ	enc	e k	ind	: m	RNA									•		
	Orig	ina	1 s	ourc	е												
	О	rga	nisı	n :	mou	se											
15	Sequ	enc	e cl	hara	cte	rist	ics										
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20	S	Symb	01-	expr	ess	ing	cha	rac	teri	isti	cs	: si	g p	ept:	ide		
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	Val	Met	Thr		Ser	His	Lys	Phe		Ser	Thr	Ser	Val		Asp	Arg	•
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35																	
Sequence length: 420 Sequence type: nucleic acid Strandedness: double Topology: linear Sequence kind: mRNA Original source Organism: mouse Sequence characteristics Symbol expressing characteristics: CDS Presence position: 31420 Characteristics determination method: S Symbol expressing characteristics: sig peptide Presence position: 3190 Characteristics determination method: S Sequence characteristics: CDS Sequence Sequence characteristics Sequence Se																	
•																	240
	Trp			Gln	Lys	Pro		Gln	ser	Pro	Lys		reu	Leu	чут	ser	
40	GCA			CGG	TAC	ACT		GTC	CCT	GAT	CAC		ACT	GGC	AGT	GGA	288
																	336
45	ser	GIY	Int	ASP		THE	riie	1111	116			vai	Gili	Λια		тэр	
	CTG	GCA	GTT	TAT		TGT	CAG	CAA	CAT	TAT	AGT	CCT	CCT	CTC	ACG	TTC	384
	Leu	Ala	Val	Tyr	Tyr	Cys	Gln	Gln		Туг	Ser	Pro	Pro		Thr	Phe	
<b>E</b> 0							<b>0.1.</b>	<b>~~</b> ~		~~~		~~~		95			420
ου																	420
	GIY	.,,,,		1111	ny 2	_cu			_, -	3							

	Sequence Listing	
5	Seq. I.D. number : 10	
•	Sequence length: 360	
	Sequence type : nucleic acid	
	Strandedness : double	
10	Topology : linear	
•	Sequence kind: mRNA	
	Original source	
	Organism : mouse	
15	Sequence characteristics	
	Symbol expressing characteristics : CDS	
	Presence position: 1360	
20	Characteristics determination method: S	
	Symbol expressing characteristics : sig peptide	
	Presence position : 112	
	Characteristics determination method : S	
25	Sequence	
•	GGT GTT GAC GGA GAC ATT GTG ATG ACA CAG TCT CAC AAA TTC ATG TCC	
	Gly Val Asp Gly Asp Ile Val Met Thr Gln Ser His Lys Phe Met Ser	48
20	1 . 5 ₁₀	
30	ACA TCA GTT GGA GAC AGG GTC ACC ATC ACC TGC AAG GCC AGT CAG GAT	96
	Thr Ser Val Gly Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asp	
	GTG ACT ACT GAT GTA GCC TGG TAT CAA CAG AAA CCA CGA CAA TCT CCT	
35	Val Thr Thr Asp Val Ala Trp Tyr Gin Gln Lys Pro Arg Gln Ser Pro	144
	30 35 40	
	AAA CTA CTG ATT TAC TCG GCA TCC TAT CGG TAC ACT GGA GTC CCT GAT	192
	Lys Leu Leu Ile Tyr Ser Ala Ser Tyr Arg Tyr Thr Gly Val Pro Asp 45 50 55	
40	CGC TTC ACT GGC AGT GGA TCT GGG ACG GAT TTC ACT TTC ACC ATC AGC	240
	Arg Phe Thr Gly Ser Gly Ser Gly Thr Asp Phe Thr Phe Thr Ile Ser	240
	60 65 70 75	
	AGT GTG CAG GCT GAA GAC CTG GCA GTT TAT TAC TGT CAG CAA CAT TAT	288
45	Ser Val Gln Ala Glu Asp Leu Ala Val Tyr Tyr Cys Gln Gln His Tyr	
	AGT ACT GCG TGG ACG TTC GGT GGT GGC ACC AAG CTG GAA ATC AAA CGG	226
	Ser Thr Ala Trp Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys Arg	336
50	95 100 105	
50	GCT GAT GCT GCA CCA ACT GTA TCC	360
	Ala Asp Ala Ala Pro Thr Val Ser	
	110 115	

-

# SEQUENCE LISTING

5	(1) GENERAL INFORMATION:	
	(i) APPLICANT:	
1Ò	<ul> <li>(A) NAME: HAGIWARA, Yoshihide</li> <li>(B) STREET: 4-14, Hiraisanso</li> <li>(C) CITY: Takarazuka-shi</li> <li>(D) STATE: Hyogo-ken</li> <li>(E) COUNTRY: Japan</li> <li>(F) POSTAL CODE (ZIP): none</li> </ul>	
15	(ii) TITLE OF INVENTION: AMINO ACID SEQUENCES OF ANTI-IDIOTY: ANTIBODIES AGAINST ANTI-CANCER HUMAN MONOCLONAL ANTIBODIES AND DNA BASE SEQUENCES ENCODING THOSE SEQUENCES	PIC DY,
20	(iii) NUMBER OF SEQUENCES:48	
25	(iv) COMPUTER READABLE FORM:  (A) MEDIUM TYPE:Floppy disk  (B) COMPUTER:IBM PC compatible  (C) OPERATING SYSTEM:MS DOS 4.0  (D) SOFTWARE:Microsoft Word, Version 5.5	•
	(v) CURRENT APPLICATION DATA:  (A) APPLICATION NUMBER: EP 94 115 683.8  (B) FILING DATE: October 5, 1994	
30	(2) INFORMATION FOR SEQ ID NO: 1:	
35	(i) SEQUENCE CHARACTERISTICS:  (A) LENGTH:5 amino acids  (B) TYPE:amino acid  (D) TOPOLOGY:linear	
	<pre>(ii) MOLECULE TYPE:protein (ix) FEATURE:</pre>	
40	(xi) SEQUENCE DESCRIPTION: SEQ ID NO:1:	
	Ser Tyr Trp Met His 5	
45	(2) INFORMATION FOR SEQ ID NO: 2:	
	(i) SEQUENCE CHARACTERISTICS:  (A) LENGTH:5 amino acids  (B) TYPE:amino acid  (D) TOPOLOGY:linear	
50	<pre>(ii) MOLECULE TYPE:protein (ix) FEATURE:</pre>	
	(D) OTHER INFORMATION: hypervariable region (xi) SEQUENCE DESCRIPTION: SEQ ID NO: 2:	

```
Asp Tyr Tyr Met Asn
5
               INFORMATION FOR SEQ ID NO: 3:
          (2)
          (i)
                 SEQUENCE CHARACTERISTICS:
                   (A) LENGTH:5 amino acids
                   (B)
                        TYPE:amino acid
                   (D) TOPOLOGY:linear
10
                MOLECULE TYPE:protein
         (ii)
         (ix)
                 FEATURE:
                   (A) NAME/KEY:H-CDR1-3
                      OTHER INFORMATION: hypervariable region
                   (D)
         (xi)
                 SEQUENCE DESCRIPTION: SEQ ID NO: 3:
15
         Asn Tyr Trp Met Gln
         (2)
              INFORMATION FOR SEQ ID NO: 4:
20
                SEQUENCE CHARACTERISTICS:
         (i)
                   (A) LENGTH: 17 amino acids
                   (B)
                       TYPE:amino acid
                   (D) TOPOLOGY: linear
                MOLECULE TYPE:protein
         (ii)
25
         (ix)
                FEATURE:
                   (A) NAME/KEY:H-CDR2-1
                  (D) OTHER INFORMATION: hypervariable region
         (xi)
                SEQUENCE DESCRIPTION: SEQ ID NO: 4:
         Ala Ile Tyr Pro Gly Asn Ser Asp Ile Ser Tyr Ser Gln Asn Phe Lys
30
         Asp
              INFORMATION FOR SEQ ID NO: 5:
         (2)
         (i)
                SEQUENCE CHARACTERISTICS:
35
                  (A) LENGTH: 19 amino acids
                  (B) TYPE:amino acid
                  (D) TOPOLOGY: linear
         (ii)
                MOLECULE TYPE:protein
         (ix)
                FEATURE:
40
                  (A) NAME/KEY:H-CDR2-2
                  (D)
                      OTHER INFORMATION: hypervariable region
        (xi)
                SEQUENCE DESCRIPTION: SEQ ID NO:5:
        Phe Ile Arg Asn Lys Ala Asn Leu Tyr Thr Thr Asp Tyr Ser Ala Ser
45
        Val Lys Gly
        (2)
             INFORMATION FOR SEQ ID NO: 6:
        (i)
                SEQUENCE CHARACTERISTICS:
                  (A) LENGTH: 19 amino acids
                      TYPE:amino acid
                  (D) TOPOLOGY: linear
        (ii)
               MOLECULE TYPE:protein
        (ix)
               FEATURE:
```

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	(A) NAME/RET:H-CDR2-3 (D) OTHER INFORMATION:hypervariable region
5	(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 6:
	Phe Ile Arg Asn Lys Ala Asn Tyr Tyr Thr Thr Glu Tyr Ser Ala Ser
10	Val Lys Gly
	(2) INFORMATION FOR SEQ ID NO: 7:
15	<ul> <li>(i) SEQUENCE CHARACTERISTICS: <ul> <li>(A) LENGTH:17 amino acids</li> <li>(B) TYPE:amino acid</li> <li>(D) TOPOLOGY:linear</li> </ul> </li> <li>(ii) MOLECULE TYPE:protein</li> <li>(ix) FEATURE:</li> </ul>
20	(A) NAME/KEY:H-CDR2-4 (D) OTHER INFORMATION:hypervariable region (xi) SEQUENCE DESCRIPTION:SEQ ID NO:7:
	Ala Ile Tyr Pro Gly Asp Gly Asp Thr Arg Tyr Thr Glu Lys Phe Lys
	5 10 15 Gly
25	(2) INFORMATION FOR SEQ ID NO: 8:
	(i) SEQUENCE CHARACTERISTICS:  (A) LENGTH: 10 amino acids  (B) TYPE: amino acid
30	(D) TOPOLOGY:linear (ii) MOLECULE TYPE:protein (ix) FEATURE: (A) NAME/KEY:H-CDR3-1
	(D) OTHER INFORMATION: hypervariable region (xi) SEQUENCE DESCRIPTION: SEQ ID NO: 8:
35	Glu Glu Tyr Asp Tyr Asp Thr Leu Asp Tyr 5 10
	(2) INFORMATION FOR SEQ ID NO: 9:
40	<ul> <li>(i) SEQUENCE CHARACTERISTICS:         <ul> <li>(A) LENGTH:11 amino acids</li> <li>(B) TYPE:amino acid</li> <li>(D) TOPOLOGY:linear</li> </ul> </li> </ul>
45	<pre>(ii) MOLECULE TYPE:protein (ix) FEATURE:</pre>
•	(D) OTHER INFORMATION: hypervariable region (xi) SEQUENCE DESCRIPTION: SEQ ID NO:9:
50	Asp Arg Gly Gly Arg Asp Trp Tyr Phe Asp Val 5 10
	(2) INFORMATION FOR SEQ ID NO: 10:
	(i) SEQUENCE CHARACTERISTICS:
<i>5</i> 5	

```
(A) LENGTH: 11 amino acids
                   (B) TYPE:amino acid(D) TOPOLOGY:linear
 5
          (ii)
                 MOLECULE TYPE:protein
         (ix)
                 FEATURE:
                   (A) NAME/KEY:H-CDR3-3(D) OTHER INFORMATION:hypervariable region
         (xi)
                 SEQUENCE DESCRIPTION: SEQ ID NO: 10:
 10
         Asp Gly Phe Leu Arg Asp Trp Tyr Phe Asp Val
                           5
              INFORMATION FOR SEQ ID NO: 11:
         (2)
15
         (i)
                SEQUENCE CHARACTERISTICS:
                   (A) LENGTH: 12 amino acids
                   (B) TYPE:amino acid
                   (D) TOPOLOGY:linear
         (ii)
                MOLECULE TYPE:protein
20
         (ix)
                FEATURE:
                   (A) NAME/KEY:H-CDR3-4
                   (D) OTHER INFORMATION: hypervariable region
         (xi)
                SEQUENCE DESCRIPTION: SEQ ID NO:11:
         Ser Gly Tyr Tyr Gly Ser Phe Val Gly Phe Ala Tyr
25
         (2)
              INFORMATION FOR SEQ ID NO: 12:
         (i)
                SEQUENCE CHARACTERISTICS:
30
                  (A) LENGTH:17 amino acids
                  (B)
                      TYPE:amino acid
                  (D)
                      TOPOLOGY: linear
         (ii)
                MOLECULE TYPE:protein
        (ix)
                FEATURE:
                  (A) NAME/KEY:L-CDR1-1
35
                      OTHER INFORMATION: hypervariable region
        (xi)
                SEQUENCE DESCRIPTION: SEQ ID NO: 12:
        Tyr Arg Ala Ser Lys Ser Val Gln Leu His Leu Ala Ile Val Tyr Met
                                                                    15
40
        His
        (2)
             INFORMATION FOR SEQ ID NO: 13:
                SEQUENCE CHARACTERISTICS:
        (i)
                  (A) LENGTH: 16 amino acids
45
                  (B) TYPE:amino acid
                     TOPOLOGY:linear
                  (D)
               MOLECULE TYPE:protein
        (ii)
        (ix)
               FEATURE:
                  (A) NAME/KEY:L-CDR1-2
50
                  (D) OTHER INFORMATION: hypervariable region
               SEQUENCE DESCRIPTION: SEQ ID NO:13:
        (xi)
        Tyr Arg Ala Ser Lys Ser Val Ser Thr Ser Gly Tyr Ser Tyr Met His
```

	(2) INFORMATION FOR SEQ ID NO: 14:
5	(i) SEQUENCE CHARACTERISTICS:  (A) LENGTH:11 amino acids  (B) TYPE:amino acid  (D) TOPOLOGY:linear
10	<pre>(ii) MOLECULE TYPE:protein (ix) FEATURE:</pre>
	(D) OTHER INFORMATION: hypervariable region (xi) SEQUENCE DESCRIPTION: SEQ ID NO: 14:
15	Lys Ala Ser Gln Asp Val Asn Thr Ala Val Ala 5 10
	(2) INFORMATION FOR SEQ ID NO: 15:
20	<ul> <li>(i) SEQUENCE CHARACTERISTICS:         <ul> <li>(A) LENGTH:11 amino acids</li> <li>(B) TYPE:amino acid</li> <li>(D) TOPOLOGY:linear</li> </ul> </li> </ul>
	(ii) MOLECULE TYPE:protein (ix) FEATURE: (A) NAME/KEY:L-CDR1-4
<b>25</b>	(D) OTHER INFORMATION: hypervariable region (xi) SEQUENCE DESCRIPTION: SEQ ID NO: 15:
	Lys Ala Ser Gln Asp Val Thr Thr Asp Val Ala 5 10
30	(2) INFORMATION FOR SEQ ID NO: 16:
<i>35</i>	(i) SEQUENCE CHARACTERISTICS:  (A) LENGTH: 7 amino acids  (B) TYPE: amino acid  (D) TOPOLOGY: linear
	<pre>(ii) MOLECULE TYPE:protein (ix) FEATURE:</pre>
40	(A) NAME/KEY:L-CDR2-1 (D) OTHER INFORMATION: hypervariable region (xi) SEQUENCE DESCRIPTION: SEQ ID NO: 16:
	Leu Val Ser Asn Leu Glu Ser 5
45	(2) INFORMATION FOR SEQ ID NO: 17:
	(i) SEQUENCE CHARACTERISTICS:  (A) LENGTH:7 amino acids  (B) TYPE:amino acid  (D) TOPOLOGY:linear
50	<pre>(ii) MOLECULE TYPE:protein (ix) FEATURE:</pre>
	(D) OTHER INFORMATION: hypervariable region (xi) SEQUENCE DESCRIPTION: SEQ ID NO: 17:

```
Leu Val Ser Asn Leu Asp Ser
                    INFORMATION FOR SEQ ID NO: 18:
               (2)
                      SEQUENCE CHARACTERISTICS:
               (i)
                         (A) LENGTH: 7 amino acids
                         (B)
                             TYPE:amino acid
10
                         (D) TOPOLOGY: linear
                      MOLECULE TYPE:protein
               (ii)
               (ix)
                      FEATURE:
                         (A)
                             NAME/KEY:L-CDR2-3
                             OTHER INFORMATION: hypervariable region
                         (D)
                      SEQUENCE DESCRIPTION: SEQ ID NO: 18:
               (xi)
15
               Ser Ala Ser Tyr Arg Tyr Thr
               (2)
                    INFORMATION FOR SEQ ID NO: 19:
20
               (i)
                      SEQUENCE CHARACTERISTICS:
                        (A) LENGTH:8 amino acids
                        (B)
                            TYPE:amino acid
                            TOPOLOGY: linear
                        (D)
               (ii)
                      MOLECULE TYPE:protein
25
               (ix)
                      FEATURE:
                            NAME/KEY:L-CDR3-1
OTHER INFORMATION:hypervariable region
                        (A)
               (xi)
                      SEQUENCE DESCRIPTION: SEQ ID NO:19:
              Gln His Ile Arg Val Ala Tyr Thr
30
              (2) INFORMATION FOR SEQ ID NO: 20:
              (i)
                      SEQUENCE CHARACTERISTICS:
                            LENGTH:8 amino acids
                            TYPE:amino acid
                        (D) TOPOLOGY: linear
              (ii)
                     MOLECULE TYPE:protein
              (ix)
                      FEATURE:
                        (A) NAME/KEY:L-CDR3-2
40
                             OTHER INFORMATION: hypervariable region
                        (D)
              (xi)
                      SEQUENCE DESCRIPTION: SEQ ID NO: 20:
              Gln His Ile Arg Gly Ala Tyr Thr
45
              (2)
                   INFORMATION FOR SEQ ID NO: 21:
              (i)
                     SEQUENCE CHARACTERISTICS:
                        (A) LENGTH:8 amino acids
                             TYPE:amino acid
                        (B)
                        (D)
                             TOPOLOGY: linear
50
                     MOLECULE TYPE:protein
              (ii)
              (ix)
                     FEATURE:
                        (A)
                             NAME/KEY:L-CDR3-3
                        (D)
                             OTHER INFORMATION: hypervariable region
```

```
(xi)
               SEQUENCE DESCRIPTION: SEQ ID NO:21:
        Gln His Ile Glu Gly Ala Tyr Thr
        (2)
             INFORMATION FOR SEQ ID NO: 22:
               SEQUENCE CHARACTERISTICS:
        (i)
10
                 (A) LENGTH:9 amino acids
                 (B) TYPE:amino acid
                 (D) TOPOLOGY: linear
               MOLECULE TYPE:protein
        (ii)
        (ix)
               FEATURE:
                 (A) NAME/KEY:L-CDR3-4
(D) OTHER INFORMATION:hypervariable region
15
               SEQUENCE DESCRIPTION: SEQ ID NO: 22:
       (xi)
       Gln Gln His Tyr Ser Pro Pro Leu Thr
                         5
20
       (2)
             INFORMATION FOR SEQ ID NO: 23:
               SEQUENCE CHARACTERISTICS:
       (i)
                 (A) LENGTH: 9 amino acids
                 (B)
                      TYPE:amino acid
                 (D) TOPOLOGY: linear
25
       (ii)
              MOLECULE TYPE:protein
       (ix)
               FEATURE:
                 (A) NAME/KEY:L-CDR3-5
                 (D) OTHER INFORMATION: hypervariable region
       (xi)
              SEQUENCE DESCRIPTION: SEQ ID NO:23:
30
       Gln Gln His Tyr Ser Thr Ala Trp Thr
       (2)
            INFORMATION FOR SEQ ID NO: 24:
35
              SEQUENCE CHARACTERISTICS:
       (i)
                 (A) LENGTH: 34 base pairs
                 (B) TYPE:nucleic acid
                 (C) STRANDEDNESS:single
(D) TOPOLOGY:linear
       (ii)
              MOLECULE TYPE: CDNA
40
       (iv)
              ANTISENSE: no
       (iii)
              HYPOTHETICAL: no
       (ix)
              FEATURE:
                 (A) NAME/KEY: H Leader Sequence A
                 (D) OTHER INFORMATION: R is A or G:
45
                                          S is C or G;
                                          K is G or T;
                                          Y is C or T;
                                          M is A or C.
       (xi)
              SEQUENCE DESCRIPTION: SEQ ID NO: 24:
50
       GGGAATTCAT GRASTTSKGG YYTMARCTKG RTTT
                                                                               34
       (2)
            INFORMATION FOR SEO ID NO: 25:
       (i)
              SEQUENCE CHARACTERISTICS:
```

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5	(B) TYPE:nucleic acid (C) STRANDEDNESS:single (D) TOPOLOGY:linear	
	(ii) MOLECULE TYPE:cDNA (iii) HYPOTHETICAL:no (iv) ANTISENSE:no	
10	(ix) FEATURE:  (A) NAME/KEY:H Leader Sequence B  (D) OTHER INFORMATION:S is C or G;  Y is C or T;  W is A or T;  R is A or G.	
15	(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 25:	
	GGGAATTCAT GRAATGSASC TGGGTYWTYC TCTT	` 34
	(2) INFORMATION FOR SEQ ID NO: 26:	
20	(i) SEQUENCE CHARACTERISTICS: (A) LENGTH:18 base pairs (B) TYPE:nucleic acid	
25	(C) STRANDEDNESS:single (D) TOPOLOGY:linear (ii) MOLECULE TYPE:cDNA (iii) HYPOTHETICAL:no	
23	(iv) ANTISENSE:no (ix) FEATURE:	
•	(A) NAME/KEY:H Leader Sequence C (xi) SEQUENCE DESCRIPTION:SEQ ID NO: 26:	
30	TTAAATGGTA TCCAGTGT	18
	(2) INFORMATION FOR SEQ ID NO: 27:	
3 <i>5</i>	<ul><li>(i) SEQUENCE CHARACTERISTICS:</li><li>(A) LENGTH:35 base pairs</li><li>(B) TYPE:nucleic acid</li></ul>	
	(C) STRANDEDNESS:single (D) TOPOLOGY:linear (ii) MOLECULE TYPE:cDNA	
10	(iii) HYPOTHETICAL:no (iv) ANTISENSE:no (ix) FEATURE:	
	(A) NAME/KEY:H Constant Region (D) OTHER INFORMATION:R is A or G; K is G or T; N is inosine.	
15	(xi) SEQUENCE DESCRIPTION:SEQ ID NO:27:	
	CCCAAGCTTC CAGGGRCCAR KGGATARACN GRTGG	35
: <b>o</b>	(2) INFORMATION FOR SEQ ID NO: 28: (i) SEQUENCE CHARACTERISTICS: (A) LENGTH:32 base pairs (B) TYPE:nucleic acid (C) STRANDEDNESS:single (D) TOPOLOGY:linear	

.

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	(ii) (iii)	MOLECULE TYPE:cDNA HYPOTHETICAL:no	
_	(iv)		
5	(ix)	FEATURE:	
		<ul> <li>(A) NAME/KEY:L Leader Sequence A</li> <li>(D) OTHER INFORMATION:R is A or G;</li> <li>K is G or T;</li> <li>W is A or T;</li> </ul>	
10		Y is C or T.	•
*	(xi)	SEQUENCE DESCRIPTION: SEQ ID NO: 28:	
	GGGAA'	TTCAT GRAGWCACAK WCYCAGGTCT TT	32
15	(2)	INFORMATION FOR SEQ ID NO: 29:	
22	(i)	SEQUENCE CHARACTERISTICS:  (A) LENGTH:33 base pairs  (B) TYPE:nucleic acid  (C) STRANDEDNESS:single  (D) TOPOLOGY:linear	
20	(ii)	(D) TOPOLOGY:linear MOLECULE TYPE:cDNA	
	(iii)	•	•
	(iv)		
	(ix)		
	( )	(A) NAME/KEY:L Leader Sequence B	
25	(xi)	SEQUENCE DESCRIPTION: SEQ ID NO: 29:	
	GGAAT"	TCAAT GGAGACAGAC ACACTCCTGC TAT	33
20	(2)	INFORMATION FOR SEQ ID NO: 30:	
30	<b>(1)</b>	SEQUENCE CHARACTERISTICS:	
	(-/	(A) LENGTH: 30 base pairs	
		(B) TYPE:nucleic acid	
		(C) STRANDEDNESS:single	
		(D) TOPOLOGY: linear	
35	(ii)	MOLECULE TYPE:cDNA	
	(iii)	HYPOTHETICAL: no	
-	(iv)	ANTISENSE: no	
	(ix)	FEATURE:	
		(A) NAME/KEY:L constant	
40	(xi)	SEQUENCE DESCRIPTION: SEQ ID NO: 30:	
-	CCCAA	AGCTTA CTGGATGGTG GGAAGATGGA	30
	(2)	INFORMATION FOR SEQ ID NO: 31:	
45	(i)	SEQUENCE CHARACTERISTICS:	•
		(A) LENGTH:357 base pairs (B) TYPE:nucleic acid (C) STRANDEDNESS:double	
		(D) TOPOLOGY: linear	
50	(ii)	MOLECULE TYPE:mRNA	
	(iii)		
	(iv)	ANTISENSE: no	
	(vi)	ORIGINAL SOURCE: (A) ORGANISM:mouse	
	/ i \		
	(ix)	FEATURE:	

5	(xi	.)		) N IENCE	NAME/ E DES	KEY:	Idio	3 I 1:SE(	H cha	in v	varia 31:	ble/	'Idic	17	H cl	nain	variable
•	GAG Glu	GTT Val	CAG Gln	CTC Leu	GAG Gln 5	CAG Gln	TCT Ser	GGC Gly	ACT Thr	GTG Val	CTC Leu	GCA Ala	AGG	CCI	GG( Gl ₃ 15	G GCT Ala	48
10	TCA Ser	GTG Val	AAG Lys	ATG Met 20	TCC Ser	TGC Cys	AAG Lys	GCT Ala	TCG Ser 25	GGC	TAC Tyr	ACC Thr	TTT Phe	AAC Asn 30	AGO	TAC Tyr	96
15	TGG Trp	ATG Met	CAC His 35	TGG Trp	GTA Val	AAA Lys	CAG Gln	AGG Arg 40	CCT Pro	GGA Gly	CAG Gln	GGT Gly	CTG Leu 45	GAA Glu	TGG	ATT Ile	144
	GGC	GCG Ala 50	ATT	TAT Tyr	CCT Pro	GGA Gly	AAT Asn 55	AGT Ser	GAT Asp	ATT Ile	AGC Ser	TAC Tyr 60	AGC Ser	CAG Gln	AAC Asn	TTT Phe	192
20	AAG Lys 65	GAC Asp	AGG Arg	GCC Ala	AAA Lys	CTG Leu 70	ACT Thr	GCC Ala	GTC Val	ACA Thr	TCC Ser 75	ACC Thr	AGC Ser	ACT Thr	GCC Ala	TAC Tyr 80	240
25	ATG Met	GAA Glu	CTC Leu	AGA Arg	AGC Ser 85	CTG Leu	ACA Thr	AAT Asn	GAG Glu	GAC Asp 90	TCT Ser	GCG Ala	GTC Val	TAT Tyr	TTC Phe 95	TGT Cys	288
	ACA Thr	AAA Lys	GAG Glu	GAA Glu 100	TAT Tyr	GAT Asp	TAC Tyr	GAC Asp	ACC Thr 105	CTG Leu	GAC Asp	TAC Tyr	TGG Trp	GGT Gly 110	CAA Gln	GGA Gly	336
30	ACC Thr	Ser	GTC Val 115	ACC Thr	GTC Val	TCC Ser	TCA Ser										357
	(2)	INF	ORMA	TION	FOR	SEQ	ID	NO:	32:	,							
35	(i)	S	EQUE: (A) (B) (C) (D)	LE: TY: ST:	NGTH PE:n RAND	:366 ucle EDNE	RIST basic a ic a SS:d inea	e pa cid oubl	irs			•					
40 _.	(ii) (iii) (iv) (vi)	H: Al	OLECT YPOTI NTISI RIGII (A)	ULE : HETI ENSE NAL :	TYPE CAL:: :no SOUR	:mRN	<b>A</b>	-									
	(ix)	F	EATUI (A)	RE:				30 17				L 9 _ ·					
45	(xi)	SI		NCE I	DESCI	RIPT	ION:	SEQ	cha. ID N	D: 3	arıa: 2:	pTG					
50 ·	GAG G	TG A	AAG ( Lys I	CTG ( Leu V	GTG ( Val (	GAG 2 Glu S	CT ( Ser (	GGA (	Gly (	GGC T Gly 1	rtg ( Leu 1	GTA ( Val (	CAG ( Gln )	Pro (	GGG Gly 15	GGT Gly	48
	TCT C	TC A eu A	red r	TC T Leu S	CC Ter C	TGT (	SCA A Ala T	hr s	TCT ( Ser ( 25	GGG T	TTA / Leu 1	ACC Thr I	Phe 1	ACT (Thr 1	SAT '	TAC Tyr	96

5	TAC	ATC	AAC Asn 35	TGG Trp	GTC Val	CGC Arg	CAG Gln	CCT Pro 40	CCA Pro	GGA Gly	AAG Lys	GAA Glu	CTT Leu 45	GAA Glu	TGG Trp	TTG Leu	144
·	GGT Gly	TTI Phe 50	ATT	AGA Arg	AAC Asn	AAA Lys	GCT Ala 55	AAT Asn	CTT Leu	TAC Tyr	ACA Thr	ACA Thr 60	GAC Asp	TAC Tyr	AGT Ser	GCA Ala	192
10	TCT Ser 65	GTG Val	AAG Lys	GGT Gly	CGG Arg	TTC Phe 70	ACC Thr	ATC Ile	TCC Ser	AGA Arg	CAT Asp 75	AAT Asn	CCC Pro	CAA Gln	AGC Ser	ATC Ile 80	240
15	CTC Leu	TAT Tyr	CTT Leu	CAA Gln	ATG Met 85	AAC Asn	ACC Thr	CTG Leu	ACA Thr	ACT Thr 90	GAG Glu	GAC Asp	AGT Ser	GCC Ala	ACT Thr 95	TAT Tyr	288
	TAC Tyr	TGT Cys	GCA Ala	AGA Arg 100	GAT Asp	AGG Arg	GGG Gly	GGG Gly	AGG Arg 105	GAC Asp	TGG Trp	TAC Tyr	TTC Phe	GAT Asp 110	GTC Val	TGG Trp	336
20							ACC Thr										366
<b>25</b>	(2) (i)	IN	FORMI SEQUI (A) (B) (C)	ENCE LE TY ST	CHAF NGTH PE:r RANI	ACTE 1:366 ucle EDNE	ID RIST bas ic a SS:d	ICS: e pa cid loubl	irs								
30	(ii (ii (iv (vi (ix)	L) 1	MOLEC HYPOT ANTIS ORIGI (A) FEATU	CULE THETI SENSE INAL OF JRE:	TYPE CAL: : no SOUR GANI	:mRN no CE:	iA iouse	<b>.</b>									
35	(xi	) :	(A) SEQUE				dio 'ION:					ble			-		
							TCT Ser										48
40							GCA Ala						Phe				96
<b>4</b> 5							CAG Gln					Ala					144
	GGT Gly	TTT Phe 50	ATT Ile	AGA Arg	AAC Asn	Lys	GCT Ala 55	AAT Asn	TAT Tyr	TAC Tyr	Thr	ACA Thr 60	GAG Glu	TAC Tyr	AGT Ser	GCA Ala	192

•

50

<b>5</b>	TC Se: 65	T 40	MG AA	G GG s Gl	r CGG	TTC Phe 70	ACC Thr	C ATO	C TC	C AG	A GAT g Asp 75	raa 1 Taa c	TCC Ser	CA#	A AGC	ATC Ile 80	240
	CT( Le	C TA	T CT n Me	T CAI t Asi	A ATO Thr 85	AAC Leu	ACC Thr	CTC Let	3 AGA	A GCT g Ala 90	GAG Glu	GAC Asp	AGT Ser	GCC	ACT Thr 95	TAT	288
10	TA(	TG Cy	T GC	A AGA A Arg 100	, Asp	GGG Gly	TTC Phe	CTA Leu	A CGG Arg 105	j Ast	TGG Trp	TAC Tyr	TTC Phe	GAT Asp 110	Val	TGG Trp	336
15	GG( Gly	C GC.	A GGC a Gly 115	G ACC 7 Thr	ACG Thr	GTC Val	ACC Thr	Val	Ser	TCA Ser	•						366
	(2)	I	NFORM	(ATIO	N FO	R SE	Q ID	NO:	34:								
20	(i)		SEQU (A (E (C	3) T :) S	CHA ENGT YPE: TRAN	H:36 nucle DEDNI	3 bas eic a ESS:	se p acid doub	airs	٠							٠.
	(ii (ii	•	MOLE	CULE THET	TYP	E:mRI		ar.									
25	(iv (vi	) ်	ANTI	SENS INAL	E:no	RCE:	M01154		٠.								
	(ix	)	FEAT	URE:	AME/I				H ch	ain '	varia	ahla		•			
30	(xi	)		ÉNCE													
	GAG Glu	GTT Val	CAG Gln	CTC	CAG Gln 5	CAG Gln	TCT Ser	GGG Gly	GCT Ala	GAA Glu 10	CTG Leu	GCA Ala	AGA Arg	CCT Pro	GGG Gly 15	GCT Ala	48
35	TCA Ser	GTG Val	AAC Asn	TTG Leu 20	TCC Ser	TGC Cys	AAG Lys	GCT Ala	TCT Ser 25	GGC Gly	TAC Tyr	ACC Thr	TTT Phe	ACT Thr 30	AAC Asn	TAC Tyr	96
<b>4</b> 0	TGG Trp	ATG Met	CAG Gln 35	TGG Trp	GTA Val	AAA Lys	CAG Gln	AGG Arg 40	CCT Pro	GGA Gly	CAG Gln	GGT Gly	CTG Leu 45	GAA Glu	TGG Trp	ATT Ile	144
	GGG Gly	GCT Ala 50	ATT	TAT Tyr	CCT Pro	GGA Gly	GAT Asp 55	GGT Gly	GAT Asp	ACT Thr	AGG Arg	TAC Tyr 60	ACT Thr	CAG Gln	AAG Lys	TTC Phe	192
45	AAG Lys 65	GGC Gly	AAG Lys	GCC Ala	ACA Thr	TTG Leu 70	ACT Thr	GCA Ala	GCT Ala	AAA Lys	TCC Ser 75	TCC Ser	AGC Ser	ACA Thr	Ala	TAC Tyr 80	240
50	ATG Met	CAA Gln	CTC Leu	AGC Ser	AGC Ser 85	TTG Leu	GCA Ala	TCT Ser	GAG Glu	GAC Asp 90	TCT Ser	GCG Ala	GTC Val	TAT Tyr	TAC Tyr 95	TGT Cys	288

<i>5</i> .	GCA Ala	AGA	TCG Ser	GGC Gly 100	Tyr	TAT Tyr	GGT Gly	AGC Ser	TTC Phe 105	Val	GGG Gly	TTT Phe	GCT Ala	TAC Tyr 110	TGG Trp	GCC	336
			ACT Thr 115														363
10	(2)	IN	FORM	ATIO	N FO	R SE	מז מ	NO:	35:								
	(i)		SEQU (A	ENCE	CHA		ERIS'	TICS	:			٠				,	
15			•	) S' ) T(	TRANI OPOL	DEDNI DGY:	ESS:	doub.	le								
	(ii (iv	i) )	MOLE: HYPO' ANTI:	THET: SENS!	ICAL E:no	: no	NA										
20	(vi (ix	•	ORIG (A FEAT	) OI	RGAN:	ISM:			ah a			-1-				. •	•
	(xi	)	( A SEQUI								arial 35:	oře.					
25			GTG Val														48
30			AGG Arg														96
			GCT Ala 35														144
			AGA Arg														192
40			AGG Arg														240
			CCT														288
45			GTA Val														336
50	(2)	IN	FORM	MOITA	I FOF	R SEC	) ID	NO:	36:								
	(i)	:	SEQUE	NCE	CHAF	ACTE	RIST	rics:									

			(B	•			eic a										
			(C	: _			ESS:		le								
5	(ii	`	( D MOLE	•			line	ar									
	(ii:	,	HYPO!				M					•					
	(iv		ANTI			. 110											
	(vi		ORIG			ocr.			•								
	( • = .	,	(A)				nouse	<b>.</b>									
10	(ix)	١ .	FEAT	•				_									
	<b>\</b>	,	(A)		AME/I	KEY:	Idio	17 1	L ch	ain v	varia	able					
	(xi)	)	SEQUI	•							_						
	`																
	GAC	ATT	GTG	CTG	ACA	CAG	TCT	CCT	GCT	TCC	TTA	GCT	GTA	TCT	CTG	GGG	48
			Val														
15	-				5					10					15	•	
			GCC														96
	Gln	Arg	Ala	Ser	Ile	Ser	Tyr	Arg	Ala	Ser	Lys	Ser	Val	Ser	Thr	Ser	
				20					25					30			
20																	
			AGT														144
	Gly	Tyr	Ser	Tyr	Met	His	Trp		GIn	Gln	Lys	Pro		Gln	Pro	Pro	٠.,
			35					40					45				
			om o							O.T. \		mari	000	ama	00m		100
			CTC														192
25	Arg		Leu	TIE	TYL	Leu		Ser	ASII	ren	GIU		GIA	vai	PIO	WIG	
		50					55					60					
	NCC.	THE	AGT	ccc	y Cut	ccc	ጥርጥ	ccc	ACA	CAC	יייוירי	ACC	CTC	270	ልጥሮ	СЪТ	240
			Ser														240
	65	1 110	001	GIJ	561	70		0-1	4		75					80	
30	05					, ,											
	CCT	GTG	GAG	GAG	GAG	GAT	GCT	GCA	ACC	TAT	TAC	TGT	CAG	CAC	ATT	AGG	288
			Glu														
					85	•				90	•	-			95	•	
35			TAC														330
33	Gly	Ala	Tyr	Thr	Phe	Gly	Gly	Gly	Thr	Lys	Leu	Glu	Ile	Lys	,		
				100					105					110			
																	-
	(2)	IN	FORMA	ATION	I FOF	₹ SEÇ	DI	NO:	37:								
40																	
	(i)		SEQUE														
			(A)				) bas		ills				•				
			(B)				eic a ESS:c		١.								
			(C) (D)				linea		. =								
45	/;; \	. 1	MOLEC					A.L.									
	(ii) (iii		HYPOI				173						•				
	(iv)	-	ANTIS			110											
	(vi)		ORIGI			CF.											
	( ~ ~ )	`	(A)				ouse	<b>.</b>									
	(ix)	, 1	FEATU		·21111]	1-1 H		•									
50	( /		(A)	NZ	ME/F	(EY : 1	dio	20 T	cha	in v	aria	able					
	(xi)		SEQUE	ENCE	DESC	RIPT	ION:	SEO	ID N	10:	37:						
	·/	•				4											

5	Asp	Ile	Val	Leu							Leu						70
		AGG Arg															96
10		TAT Tyr															144
15		CTC Leu 50															192
	AGG Arg 65	TTC Phe	AGT Ser	GGC Gly	AGT Ser	GGG Gly 70	TCT Ser	GGG Gly	ACA Thr	GAC Asp	TTC Phe 75	ACC Thr	CTC Leu	AAC Asn	ATC Ile	CAT His 80	240
20																GAG Glu	
25	GGA Gly	GCT Ala	TAC Tyr	ACG Thr 100	TTC Phe	GGA Gly	GGG Gly	GGG Gly	ACC Thr 105	AAG Lys	CTG Leu	GAA Glu	ATA Ile	AAA Lys 110			330
	(2)	INI	FORM	TION	I FOF	R SEQ	) ID	NO:	38:								
30	(i)	5	(A) (B) (C)	LE TY ST	ENGTH PE:1 PRANI	:321 nucle EDNE	l bas eic a ESS:c	duoi	irs					٠			
35	(ii) (ii) (iv) (vi)	L) I	(D) MOLEC HYPOT ANTIS ORIGI	CULE THET! SENSI INAL	TYPE CAL: : no SOUE	E:mRi no RCE:											
<b>40</b>	(ix)	) I	(A) JEATI (A)	JRE:			nouse [dio		L cha	ain v	varia	able					
	(xi	) SI						SEQ I									
45	GAC Asp	ATT Ile	GTG Val	ATG Met	ACC Thr 5	CAG Gln	TCT Ser	CAC His	AAA Lys	TTC Phe 10	ATG Met	TCC Ser	ACA Thr	TCA Ser	GTA Val 15	GGA Gly	48
-	GAC Asp	AGG Arg	GTC Val	AGT Ser 20	ATC Ile	ACC Thr	TGC Cys	AAG Lys	GCC Ala 25	AGT Ser	CAG Gln	GAT Asp	GTG Val	AAT Asn 30	ACT Thr	GCT Ala	96
50	GTA Val	GCC Ala	TGG Trp 35	TAT Tyr	CAA Gln	CAG Gln	AAA Lys	CCA Pro 40	GGA Gly	CAA Gln	TCT Ser	CCT Pro	AAA Lys 45	CTA Leu	CTG Leu	CTT Leu	144

5	TAC	Ser Ser 50	G GCA	A TCO	TAC Tyr	C CGG	TAC Tyr 55	ACI Thr	GG/ Gly	A GTO	C CCT	GAT ASI 60	CAC His	TTC Phe	ACT Thr	GGC	192
	AGI Ser 65	GG/ Gly	A TCT	r GG( c Gly	ACG Thr	GAT Asp 70	TTC Phe	ACT Thr	TTC Phe	ACC Thr	11e	AGC Ser	GG1 Gly	GTG Val	CAG Gln	GCT Ala 80	240
10	GAA Glu	GAC Asp	CTC Leu	GCA Ala	GTT Val 85	TAT Tyr	TAC	TGT Cys	Gln	GAA Gln 90	CAT His	TAT Tyr	AGT Ser	CCT	CCT Pro 95	CTC Leu	288
15	ACG Thr	TTC Phe	GGT Gly	GCT Ala 100	Gly	ACC Thr	AAG Lys	CTG Leu	GAA Glu 105	Leu	AAA Lys	<b>.</b>			-		321
	(2)	IN	FORM	ATIO	N FO	R SE	Q ID	NO:	39:								
<i>2</i> 0	(i)		(A (B (C	) L ) T	ENGT YPE: TRAN	RACT: H:32 nucle DEDN	l ba: eic a ESS:	se pa acid doub	airs								٠,
25	(ii (ii (iv (vi	i) }	HYPO ANTI ORIG	CULE THET SENS INAL	TYPI ICAL E:no SOUI	RCE:	NA										
	(ix)	)	A) FEAT (A)	URE:		ISM:: KEY::			. ch	ain i	vari	ahla					
30	(xi)	) ;		ENCE	DESC	CRIP	rion:	SEQ	ID	NO:	39:	uDIC					
	GAC Asp	ATT Ile	GTG Val	ATG Met	ACA Thr 5	CAG Gln	TCT Ser	CAC His	AAA Lys	TTC Phe 10	ATG Met	TCC Ser	ACA Thr	TCA Ser	GTT Val 15	GGA Gly	48
35	GAC Asp	AGG Arg	GTC Val	ACC Thr 20	ATC Ile	ACC Thr	TGC Cys	AAG Lys	GCC Ala 25	AGT Ser	CAG Gln	GAT Asp	GTG Val	ACT Thr 30	ACT Thr	GAT Asp	96
40	GTA Val	GCC Ala	TGG Trp 35	TAT Tyr	CAA Gln	CAG Gln	AAA Lys	CCA Pro 40	CGA Arg	CAA Gln	TCT Ser	CCT Pro	AAA Lys 45	CTA Leu	CTG Leu	ATT Ile	144
45	TAC Tyr	TCG Ser 50	GCA Ala	TCC Ser	TAT Tyr	CGG Arg	TAC Tyr 55	ACT Thr	GGA Gly	GTC Val	CCT Pro	GAT Asp 60	CGC Arg	TTC Phe	ACT Thr	GGC Gly	192
	AGT Ser 65	GGA Gly	TCT Ser	GGG Gly	ACG Thr	GAT Asp 70	TTC Phe	ACT Thr	TTC Phe	ACC Thr	ATC Ile 75	AGC Ser	AGT Ser	GTG Val	CAG Gln	GCT Ala 80	240
50	GAA Glu	GAC Asp	CTG Leu	GCA Ala	GTT Val 85	TAT Tyr	TAC Tyr	TGT Cys	CAG Gln	CAA Gln 90	CAT His	TAT Tyr	AGT Ser	ACT Thr	GCG Ala 95	TGG Trp	288

5										ATC Ile							321
10	(2) (i)			) TY	CHAI ENGTI YPE : 1 TRANI	RACTE 1:399 nucle DEDNE		rics: se pa acid doubl	: airs				·				
15	(ii) (ii) (iv) (vi)	Ĺ) · l ) · i	HYPOT ANTI:	CULE THET: SENSI INAL ) OF	CAL: E:no SOUI	no RCE:	NA nouse	9	-								
20	(ix)	•	FEATU (A) SEQUI	) N2			Clone			10:40	):						
	CTG Leu	TCG Ser	GTA Val	ACT Thr	TCA Ser -5	GGG Gly	GTC Val	TAC Tyr	TCA Ser	GAG Glu 1	GTT Val	CAG Gln	CTC Leu	GAG Gln 5	CAG Gln	TCT · Ser	-48
25	GGG Gly	ACT Thr	GTG Val 10	CTG Leu	GCA Ala	AGG Arg	CCT Pro	GGG Gly 15	GCT Ala	TCA Ser	GTG Val	AAG Lys	ATG Met 20	TCC	TGC Cys	AAG Lys	96
30	GCT Ala	TCG Ser 25	GGC Gly	TAC Tyr	ACC Thr	TTT Phe	AAC Asn 30	AGC Ser	TAC Tyr	TGG Trp	ATG Met	CAC His 35	TGG Trp	GTA Val	AAA Lys	CAG Gln	144
	AGG Arg 40	CCT Pro	GGA Gly	CAG Gln	GGT Gly	CTG Leu 45	GAA Glu	TGG Trp	ATT Ile	GGC Gly	GCG Ala 50	ATT Ile	TAT Tyr	CCT Pro	GGA Gly	AAT Àsn 55	192
35	AGT Ser	GAT Asp	ATT Ile	AGC Ser	TAC Tyr 60	AGC Ser	CAG Gln	AAC Asn	TTT Phe	AAG Lys 65	GAC Asp	AGG Arg	GCC Ala	AAA Lys	CTG Leu 70	ACT Thr	240
40	GCC Ala	GTC Val	ACA Thr	TCC Ser 75	ACC Thr	AGC Ser	ACT Thr	GCC Ala	TAC Tyr 80	ATG Met	GAA Glu	CTC Leu	AGA Arg	AGC Ser 85	CTG Leu	ACA Thr	288
	AAT Asn	GAG Glu	GAC Asp 90	TCT	GCG Ala	GTC Val	TAT Tyr	TTC Phe 95	TGT Cys	ACA Thr	AAA Lys	GAG Glu	GAA Glu 100	TAT Tyr	GAT Asp	TAC Tyr	336
45	GAC Asp	ACC Thr 105	CTG Leu	GAC Asp	TAC Tyr	TGG Trp	GGT Gly 110	CAA Gln	GGA Gly	ACC Thr	TCA Ser	GTC Val 115	ACC Thr	GTC Val	TCC Ser	TCA Ser	384
50				ACA													399

	(2)	II	VFORM	ATIC	N FC	R SE	Q ID	NO:	41:								
5	(i)	•	SEQU (A (B	) L	CHA ENGT YPE: TRAN	H:40 nucl	2 ba eic	sė p acid	airs	1							
10	(ii (ii (iv (vi	.i) ')	MOLE HYPO ANTI	) T CULE THET SENS INAL	OPOL TYP ICAL E:no SOU RGAN	OGY: E:mR :no RCE:	line NA	ar									
15	(ix (xi	•	FEAT ( A SEQU	URE: ) N	AME/	KEY:	Clon	e 17	GB7	NO:	41:						
	ATT Ile -10	Leu	TCG	GTA Val	ACT Thr	TCA Ser -5	GGG Gly	GTC Val	TAC Tyr	TCA Ser	GAG Glu 1	GTT Val	CAG Gln	CTC Leu	GAG Gln 5	CAG Gln	48
20	TCT Ser	GGG	ACT Thr	GTG Val 10	CTG Leu	GCA Ala	AGG Arg	CCT Pro	GGG Gly 15	GCT Ala	TCA Ser	GTG Val	AAG Lys	ATG Met 20	TCC Ser	TGC Cys	96
<b>25</b> -	AAG Lys	GCT Ala	TCG Ser 25	GGC Gly	TAC Tyr	ACC Thr	TTT Phe	AAC Asn 30	AGC Ser	TAC Tyr	TGG Trp	ATG Met	CAC His 35	TGG Trp	GTA Val	AAA Lys	144
30	CAG Gln	AGG Arg 40	CCT Pro	GGA Gly	CAG Gln	GGT Gly	CTG Leu 45	GAA Glu	TGG Trp	ATT Ile	GGC Gly	GCG Ala 50	ATT Ile	TAT Tyr	CCT Pro	GGA Gly	192
	AAT Asn 55	AGT Ser	GAT Asp	ATT Ile	AGC Ser	TAC Tyr 60	AGC Ser	CAG Gln	AAC Asn	TTT Phe	AAG Lys 65	GAC Asp	AGG Arg	GCC Ala	AAA Lys	CTG Leu 70	240
15	ACT Thr	GCC Ala	GTC Val	ACA Thr	TCC Ser 75	ACC Thr	AGC Ser	ACT Thr	GCC Ala	TAC Tyr 80	ATG Met	GAA Glu	CTC Leu	AGA Arg	AGC Ser 85	CTG Leu	288
o	ACA Thr	AAT Asn	GAG Glu	GAC Asp 90	TCT Ser	GCG Ala	GTC Val	TAT Tyr	TTC Phe 95	TGT Cys	ACA Thr	AAA Lys	GAG Glu	GAA Glu 100	TAT Tyr	GAT Asp	336
	TAC Tyr	GAC Asp	ACC Thr 105	CTG Leu	GAC Asp	TAC Tyr	TGG Trp	GGT Gly 110	CAA Gln	GGA Gly	ACC Thr	TCA Ser	GTC Val 115	ACC Thr	GTC Val	TCC Ser	384
5					ACA Thr												402
<b>o</b> .	(2) (i)			NCE LE	FOR CHAR NGTH	ACTE: 438	RIST bas	ICS:									•
			(0)	TI	PE:n	псте	TC 9	CIU									

5 <u>.</u>	(ii (ii (iv (vi	í) )	(C) (D) MOLE HYPO ANTI ORIG	) T CULE THET SENS INAL	OPOL TYP ICAL E:no SOU	:no RCE:	line NA	ar	le								
10	(ix (xi		(A) FEAT (A) SEQU	ÚRE: ) <b>N</b> .	AME/	ISM:: KEY:: CRIP'	Clon	e 20		NO: 4:	2:						
15	ATG Met	GAG Glu	TTC Phe	GGG Gly	CTA Leu -15	AAC Asn	TGG Trp	GTT Val	TTC Phe	CTT Leu -10	GTA Val	ACA Thr	CTT Leu	TTA Leu	AAT Asn -5	GGT Gly	48
	ATC Ile	CAG Gln	TGT Cys	GAG Glu 1	GTG Val	AAG Lys	CTG Leu	GTG Val 5	GAG Glu	TCT Ser	GGA Gly	GGA Gly	GGC Gly 10	TTG Leu	GTA Val	CAG Gln	96
20 .	CCT Pro	GGG Gly 15	GGT Gly	Ser	CTC Leu	AGA Arg	CTC Leu 20	TCC Ser	TGT Cys	GCA Ala	ACT Thr	TCT Ser 25	GGG Gly	TTA Leu	ACC Thr	TTC Phe	144
25 ·	ACT Thr 30	GAT Asp	TAC Tyr	TAC Tyr	ATG Met	AAC Asn 35	TGG Trp	GTC Val	CGC Arg	CAG Gln	CCT Pro 40	CCA Pro	GGA Gly	AAG Lys	GAA Glu	CTT Leu 45	192
	GAA Glu	TGG Trp	TTG Leu	GGT Gly	TTT Phe 50	ATT Ile	AGA Arg	AAC Asn	AAA Lys	GCT Ala 55	AAT Asn	CTT Leu	TAC Tyr	ACA Thr	ACA Thr 60	GAC Asp	240
30	TAC Tyr	AGT Ser	GCA Ala	TCT Ser 65	GTG Val	AAG Lys	GGT Gly	CGG Arg	TTC Phe 70	ACC Thr	ATC Ile	TCC Ser	AGA Arg	CAT Asp 75	AAT Asn	CCC Pro	288
35	CAA Gln	AĞC Ser	ATC Ile 80	CTC Leu	TAT Tyr	CTT Leu	CAA Gln	ATG Met 85	AAC Asn	ACC Thr	CTG Leu	ACA Thr	ACT Thr 90	GAG Glu	GAC Asp	AGT Ser	336
	GCC Ala	ACT Thr 95	TAT Tyr	TAC Tyr	TGT Cys	GCA Ala	AGA Arg 100	GAT Asp	AGG Arg	GGG Gly	GGG Gly	AGG Arg 105	GAC Asp	TGG Trp	TAC Tyr	TTC Phe	384
	_		TGG Trp							_	_						432
	ACA Thr																438
	(2)	INE	FORMA	TION	FOR	SEQ	ID	NO:	43:								
50	(i)	5	SEQUE (A) (B) (C) (D)	LE TY ST	NGTH PE:n RAND	ACTE :411 ucle EDNE	bas ic a SS:d	e pa cid oubl	irs								

5	(ii (ii (iv (vi	i)	MOLE HYPO ANTI ORIG	THET SENS INAL	ICAL E:no SOU	:no RCE:						•					
	(ix	•	(A FEAT (A	URE: ) N.	RGAN AME/	KEY:	Clon	e 27									
10	(xi	)	SEQU	ENCE	DES	CRIP	TION	: SEQ	ID	NO:	43:				•		
	CTT Leu -10	GTA Val	ACA Thr	CGT Arg	TTA Leu	AAT Asn -5	GGT Gly	ATC Ile	CAG Gln	TGT Cys	GAG Glu 1	GTG Val	AAG Lys	CTG Leu	GTG Val 5	GAG Glu	48
15	TCT Ser	GGA Gly	GGA Gly	GGC Gly 10	TTG Leu	GTA Val	CAG Gln	CCT Pro	GGG Gly 15	GGT Gly	TCT Ser	CTG Leu	AGA Arg	CTC Leu 20	TCC Ser	TGT Cys	96
20	GCA Ala	ACT Thr	TCT Ser 25	GGG Gly	TTC Phe	ACC Thr	TTC Phe	ACT Thr 30	GAT Asp	TAC Tyr	TAC Tyr	ATG Met	AAC Asn 35	TGG Trp	GTC Val	CGC Arg	144
	CAG Gln	CCT Pro 40	CCA Pro	GGA Gly	AAG Lys	GCA Ala	CTT Leu 45	GAG Glu	TGG Trp	TTG Leu	GGT Gly	TTT Phe 50	ATT Ile	AGA Arg	AAC Asn	AAA Lys	192
25	GCT Ala 55	AAT Asn	TAT Tyr	TAC Tyr	ACA Thr	ACA Thr 60	GAG Glu	TAC Tyr	AGT Ser	GCA Ala	TCT Ser 65	GTG Val	AAG Lys	GGT Gly	CGG Arg	TTC Phe 70	240
30	ACC Thr	ATC Ile	TCC Ser	AGA Arg	GAT Asp 75	AAT Asn	TCC Ser	CAA Gln	AGC Ser	ATC Ile 80	CTC Leu	TAT Gln	CTT Met	CAA Asn	ATG Thr 85	AAC Leu	288
	ACC Thr	CTG Leu	AGA Arg	GCT Ala 90	GAG Glu	GAC Asp	AGT Ser	GCC Ala	ACT Thr 95	TAT Tyr	TAC Tyr	TGT Cys	GCA Ala	AGA Arg 100	GAT Asp	GGG Gly	336
35	TTC Phe	CTA Leu	CGG Arg 105	GAC Asp	TGG Trp	TAC Tyr	TTC Phe	GAT Asp 110	GTC Val	TGG Trp	GGC	GCA Ala	GGG Gly 115	ACC Thr	ACG Thr	GTC Val	384
40	ACC	GTC Val 120	TCC Ser	TCA Ser	GCC Ala	AAA Lys	ACG Thr 125	ACA Thr	CCC Pro								411
	(2)	INI	FORMA	TION	FOR	SEC	) ID	NO:	44:								,
	(i)	5	EQUE (A) (B) (C)	LE TY ST	NGTH PE:n RAND	:354 ucle EDNE	bas ic a	e pa cid oubl	irs								•
50	(ii) (iii (iv) (vi)	) I	(D) OLEC IYPOT NTIS RIGI (A)	ULE HETI ENSE NAL	CAL:	:mRN no CE:	<b>A</b>										

	(ix	)	FEAT		AME/	KEY:	Clon	e 3K	B11								
5	(xi	)	SEQU	ENCE	DES	CRIP	TION	:SEQ	ID 1	NO:4	4:						
	GAC Asp	ATT Ile	GTG Val	CTG Leu	ACA Thr 5	CAG Gln	TCT Ser	CCT Pro	GCT Ala	TCC Ser 10	TTA Leu	GCT Ala	GTA Val	TCT Ser	CCT Pro 15	CTG Leu	48
10	GGG Gly	CAG Gln	AGG Arg	GCC Ala 20	ACC Thr	ATC Ile	TCA Ser	TAC Tyr	AGG Arg 25	GCC Ala	AGC Ser	AAA Lys	AGT Ser	GTG Val 30	CAG Gln	TTA Leu	96
15	CAT His	CTG Leu	GCT Ala 35	ATA Ile	GTT Val	TAT Tyr	ATG Met	CAC His 40	TGG Trp	AAC Asn	CAA Gln	CAG Gln	AAA Lys 45	CCA Pro	GGA Gly	CAG Gln	144
20	CCA Pro	CCC Pro 50	AGA Arg	CTC Leu	CTC Leu	ATC Ile	TAT Tyr 55	CTT Leu	GTA Val	TCC Ser	AAC Asn	CTA Leu 60	GAA Glu	TCT Ser	GGG Gly	GTC Val	192
	CCT Pro 65	GCC Ala	AGG Arg	TTC Phe	AGT Ser	GGC Gly 70	AGT Ser	GGG Gly	TCT Ser	GGG Gly	ACA Thr 75	GAC Asp	TTC Phe	ACC Thr	CTC Leu	AAC Asn 80	240
25	ATC Ile	CAT His	CCT Pro	GTG Val	GAG Glu 85	GAG Glu	GAG Glu	GAT Asp	GCT Ala	GCA Ala 90	ACC Thr	TAT Tyr	TAC Tyr	TGT Cys	CAG Gln 95	CAC His	288
30	ATT Ile	AGG Arg	GTA Val	GCT Ala 100	TAC Tyr	ACG Thr	TTC Phe	GGA Gly	GGG Gly 105	GGG Gly	ACC Thr	AAG Lys	CTG Leu	GAA Glu 110	ATA Ile	AAA Lys	336
				GCT Ala													354
35																	
	(2)	IN	ORMA	TION	FOR	SEQ	ID	NO:	45:								
40	(i)	S	SEQUE (A) (B) (C)	TY	CHAR NGTH PE: n	:438	bas	e pa	irs								
	(ii) (iii (iv)	) I	OLEC IYPOT INTIS	TO CULE CHETI SENSE	TYPE CAL:	:mRN no		r									-
45	(vi)		(A) JTAE		GANI	SM:m			В1								,
	(xi)	5		NCE						O: 4	5:				•		
50	CTA Leu	TGG Trp	GTA Val	CTG Leu -10	CTG Leu	CTC Leu	TGG Trp	Val	CCA Pro -5	GGT Gly	TCC Ser	ACT Thr	GGT Gly	GAC Asp 1	ATT Ile	GTG Val	48

5	CTG Leu	ACA Thr	CAG Gln	S TCT	CCT Pro	GCT Ala	TCC Ser 10	TTA L u	GCT Ala	GTA Val	TCT Ser	CTG Leu 15	GGG Gly	CAG Gln	AGG	GCC Ala	96
	TCC Ser 20	ATC	TCA Ser	TAC	AGG Arg	GCC Ala 25	AGC Ser	AAA Lys	AGT Ser	GTC Val	AGT Ser 30	ACA Thr	TCT Ser	GGC	TAT	AGT Ser 35	144
10	TAT Tyr	ATG Met	CAC His	TGG Trp	AAC Asn 40	CAA Gln	CAG Gln	AAA Lys	CCA Pro	GGA Gly 45	CAG Gln	CCA Pro	CCC Pro	AGA Arg	CTC Leu 50	CTC Leu	192
15	ATC Ile	TAT Tyr	CTT Leu	GTA Val 55	TCC Ser	AAC Asn	CTA Leu	GAA Glu	TCT Ser 60	GGG Gly	GTC Val	CCT Pro	GCC Ala	AGG Arg 65	TTC Phe	AGT Ser	240
	GGC Gly	AGT Ser	GGG Gly 70	TCT Ser	GGG Gly	ACA Thr	GAC Asp	TTC Phe 75	ACC Thr	CTC Leu	AAC Asn	ATC Ile	CAT His 80	CCT Pro	GTG Val	GAG Glu	288
. · ·	GAG Glu	GAG Glu 85	GAT Asp	GCT Ala	GCA Ala	ACC Thr	TAT Tyr 90	TAC Tyr	TGT Cys	CAG Gln	CAC His	ATT Ile 95	AGG Arg	GGA Gly	GCT Ala	TAC Tyr	336
25	ACG Thr 100	TTC Phe	GGA Gly	GGG Gly	GGG Gly	ACC Thr 105	AAG Lys	CTG Leu	GAA Glu	ATA Ile	AAA Lys 110	CGG Arg	GCT Ala	GAT Asp	GCT Ala	GCA Ala 115	384
30	CCA Pro	ACT Thr	GTA Val	TCC Ser	ATC Ile 120	TTC Phe	CCA Pro	CCA Pro	TCC Ser	AGT Ser 125	AAG Lys	CTT Leu	GGG Gly	AAA Lys	CGG Arg 130	TTC Phe	432
	GCA Ala																438
35	(2)	INF	ORMA	TION	FOR	SEQ	ID	NO:	46:								
	(i)	S	EQUE (A) (B) (C)	TY ST	NGTH PE:n RAND	:417 ucle EDNE:	bas ic a SS:d	e pa cid oubl									4
40	(ii) (iii (iv)	) H A	YPOT NTIS	TO ULE HETI ENSE	POLO TYPE CAL::	GY:1 :mRN no	inea	r									
<b>1</b> 5	(vi) (ix)	F	(A) EATU (A)	NAI	GANI: ME/KI	SM:mo	lone	20K	B1 [′]								
	(xi)			NCE I													
50		GG	ددودا	G GT	3AGA/	ACCG	TTG	∌GAA'.	1	ATG ( Met ( -20	GAG 1 Glu 7	ACA ( Thr 1	SAC Asp ?	Chr 1	CTC ( Leu 1 -15	CTG Leu	48

5	CTA Leu	TGG Trp	GTA Val	CTG Leu -10	CTG Leu	CTC Leu	TGG Trp	GTT Val	CCA Pro -5	GGT Gly	TCC Ser	ACT	GGT Gly	GAC Asp 1	ATT	GTG Val	
	CTG Leu	ACA Thr	CAG Gln 5	TCT Ser	CCT Pro	GCT Ala	TCC Ser	TTA Leu	GCT Ala 10	GTA Val	TCT Ser	CTG Leu	GGG Gly	CAG Gln 15	AGG Arg	GCC Ala	144
10	ACC Thr	ATC Ile	TCA Ser 20	TAC Tyr	AGG Arg	GCC Ala	AGC Ser	AAA Lys 25	AGT Ser	GTC Val	AGT Ser	ACA Thr	TCT Ser 30	GGC Gly	TAT Tyr	AGT Ser	192
15	TAT Tyr	ATG Met 35	CAC His	TGG Trp	AAC Asn	CAA Gln	CAG Gln 40	AGA Arg	CCA Pro	GGA Gly	CAG Gln	CCA Pro 45	CCC Pro	AGA Arg	CTC Leu	CTC Leu	240
	ATC Ile 50	TAT Tyr	CTT Leu	GTA Val	TCC Ser	AAC Asn 55	CTA Leu	GAC Asp	TCT Ser	GGG Gly	GTC Val 60	CCT Pro	GCC Ala	AGG Arg	TTC Phe	AGT Ser 65	288
<b>20</b>	GGC Gly	AGT Ser	GGG Gly	TCT Ser	GGG Gly 70	ACA Thr	GAC Asp	TTC Phe	ACC Thr	CTC Leu 75	AAC Asn	ATC Ile	CAT His	CCT Pro	GTG Val 80	GAG Glu	336
25							TAT Tyr										384
30							AAG Lys										417
	(2) (i)					_	ID RIST										
35		2	(A) (B) (C)	LE TY	NGTH PE: n RAND	:420 ucle EDNE	baseic a SS:s	se pa scid singl	irs								
40	(ii) (iii (iv) (vi)	.) H	IOLEÓ IYPOT NTIS RIGI	ULE THETI SENSE NAL	TYPE CAL: :no SOUR	:mRN no CE:	IA										
45	(ix)		(A) EATU (A) EQUE	JRE:	ME/K	EY:C	louse lone	27K		iO: 4	17:						
		Ģ	CGGC	CGCG	G TG	AGAA	CCGI	TTG	GGAA	TTC					TCC Ser	_	48
50							CTC Leu										96

5	Val	Met	Thr.	Gln	Ser	His	AAA Lys	Phe 10	ATG Met	TCC	ACA Thr	TCA Ser	GTA Val 15	GGA Gly	GAC Asp	AGG Arg	144
	GTC Val	AGT Ser 20	ATC	ACC	TGC Cys	AAG Lys	GCC Ala 25	AGT Ser	CAG Gln	GAT Asp	GTG Val	AAT Asn 30	ACT Thr	GCT Ala	GTA Val	GCC Ala	192
	TGG Trp 35	TAT Tyr	CAA Gln	CAG Gln	AAA Lys	CCA Pro 40	GGA Gly	CAA Gln	TCT Ser	CCT Pro	AAA Lys 45	CTA Leu	CTG Leu	CTT Leu	TAC Tyr	TCG Ser 50	240
15	GCA Ala	TCC	TAC Tyr	CGG Arg	TAC Tyr 55	ACT Thr	GGA Gly	GTC Val	CCT Pro	GAT Asp 60	CAC His	TTC Phe	ACT Thr	Gly	AGT Ser 65	GGA Gly	288
	TCT Ser	GGG Gly	ACG Thr	GAT Asp 70	TTC Phe	ACT Thr	TTC Phe	ACC Thr	ATC Ile 75	AGC Ser	GGT Gly	GTG Val	CAG Gln	GCT Ala 80	GAA Glu	GAC Asp	336
20	CTG Leu	GCA Ala	GTT Val 85	TAT Tyr	TAC Tyr	TGT Cys	CAG Gln	CAA Gln 90	CAT His	TAT Tyr	AGT Ser	CCT Pro	CCT Pro 95	CTC Leu	ACG Thr	TTC Phe	384
25	GGT Gly	GCT Ala 100	GGG Gly	ACC Thr	AAG Lys	CTG Leu	GAA Glu 105	CTG Leu	AAA Lys	CGG Arg	GCT Ala	GAT Asp 110					420
	(2)	INE	FORM	TION	FOF	SEÇ	) ID	NO:	48:								
30	(i)	S	EQUE (A) (B)	TY	NGTH PE:r	ACTE 1:360 ucle EDNE	bas	e pa	irs							•	
35	(ii) (iii (iv) (vi)	) H	(D) OLEC IYPOI NTIS	TO TULE THETI SENSE NAL	POLC TYPE CAL: :no SOUR	GY:1 :mRN no	inea A	ır	-								
40	(ix) (xi)		(A) EATU (A) EQUE	RE:	ME/K	SM:m EY:C RIPT	lone	23K		0:48							·
45	GGT	GTT Val	GAC Asp	GGA Gly	GAC Asp 1	ATT Ile	GTG Val	ATG Met	ACA Thr	CAG Gln 5	TCT Ser	CAC His	AAA Lys	TTC Phe	ATG Met 10	TCC Ser	48
	ACA Thr	TCA Ser	GTT Val	GGA Gly 15	GAC Asp	AGG Arg	GTC Val	Thr	ATC Ile 20	ACC Thr	TGC Cys	AAG Lys	GCC Ala	AGT Ser 25	CAG Gln	GAT Asp	96
50	GTG /	Thr	ACT Thr 30	GAT Asp	GTA Val	GCC Ala	Trp	TAT Tyr 35	CAA Gln	CAG Gln	AAA Lys	Pro	CGA Arg 40	CAA Gln	TCT Ser	CCT Pro	144

5			_					 CCT Pro		192
	 	 					 	 ATC Ile	AGC Ser 75	240
10	 	 						 CAT His 90		288
15								AAA Lys	CCG Arg	336
	 	 		GTA Val						360
20										

#### 25 Claims

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- 1. An immunoglobulin H chain variable region fragment which contains a hypervariable region CDR1 having an amino acid sequence selected from
  - (1) Ser Tyr Trp Met His;
    Asp Tyr Tyr Met Asn; and
    Asn Tyr Trp Met Gln;

#### a hypervariable region CDR2 having an amino acid sequence selected from

Asp Ile Tyr Pro Gly Asn Ser
Asp Ile Ser Tyr Ser Gln Asn
Phe Lys Asp;
Phe Ile Arg Asn Lys Ala
Asn Leu Tyr Thr Thr Asp
Tyr Ser Ala Ser Val Lys
Gly;
Phe Ile Arg Asn Lys Ala
Asn Tyr Tyr Thr Thr Glu
Tyr Ser Ala Ser Val Lys
Gly; and
Ala Ile Tyr Pro Gly Asp
Gly Asp Thr Arg Tyr Thr
Gln Lys Phe Lys Gly,

## and a hypervariable region CER3 having an amino acid sequenc selected in

(3) Glu Glu Tyr Asp Týr Asp Thr Leu Asp Tyr; Asp Arg Gly Gly Arg Asp Trp Tyr Phe Asp Val; Asp Gly Phe Leu Arg Asp Trp Tyr Phe Asp Val; and Ser Gly Tyr Tyr Gly Ser Phe Val Gly Phe Ala Tyr.

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## 2. An immunoglobulin H chain variable region fragment having the following amino acid sequence

20 Glu Val Gln Leu Gln Gln Ser Gly Thr Val Leu Ala Arg Pro Gly Ala Ser Val Lys Met Ser Cys Lys Ala Ser Gly Tyr Thr Phe Asn Ser Tyr Trp Met His Trp Val Lys Gln Arg 25 Pro Gly Gln Gly Leu Glu Trp Ile Gly Ala Ile Tyr Pro Gly Asn Ser Asp Ile Ser Tyr Ser Gln Asn Phe Lys Asp Arg Ala Lys Leu 30 Thr Ala Val Thr Ser Thr Ser Thr Ala Tyr Met Glu Leu Arg Ser Leu Thr Asn Glu Asp Ser Ala Val Tyr Phe Cys Thr Lys Glu Glu Tyr Asp Tyr Asp Thr Leu Asp Tyr Trp Gly Gln Gly Thr Ser Val Thr Val Ser Ser.

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#### 3. An immunoglobulin H chain variable region fragment having the following amino acid sequence

Glu Val Lys Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly Ser Leu Arg Leu Ser Cys Ala Thr Ser Gly Phe Thr Phe Thr Asp Tyr Tyr Met Asn Trp Val Arg Gln Pro Pro Gly Lys Ala Leu Glu Trp Leu Gly Phe Ile Arg Asn Lys Ala Asn Tyr Tyr Thr Thr Glu Tyr Ser Ala Ser Val Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Ser Gln Ser Ile Leu Tyr Leu Gln Met Asn Thr Leu Arg Ala Glu Asp Ser Ala Thr Tyr Tyr Cys Ala Arg Asp Gly Phe Leu Arg Asp Trp Tyr Phe Asp Val Trp Gly Ala Gly Thr Thr Val Thr Val Ser Ser.

#### 25 4. An immunoglobulin H chain variable region fragment having the following amino acid sequence

Glu Val Lys Leu Val Glu Ser Gly Gly Gly
Leu Val Gln Pro Gly Gly Ser Leu Arg Leu
Ser Cys Ala Thr Ser Gly Leu Thr Phe Thr
Asp Tyr Tyr Met Asn Trp Val Arg Gln Pro
Pro Gly Lys Glu Leu Glu Trp Leu Gly Phe
Ile Arg Asn Lys Ala Asn Leu Tyr Thr Thr
Asp Tyr Ser Ala Ser Val Lys Gly Arg Phe
Thr Ile Ser Arg Asp Asn Pro Gln Ser Ile
Leu Tyr Leu Gln Met Asn Thr Leu Thr Thr
Glu Asp Ser Ala Thr Tyr Tyr Cys Ala Arg
Asp Arg Gly Gly Arg Asp Trp Tyr Phe Asp
Val Trp Gly Ala Gly Thr Thr Val Thr Val
Ser Ser.

5. An immunoglobulin H chain variable region fragment having the following amino acid sequence

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Glu	Val	Gln	Leu	Gln	Gln	Ser	Gly	Ala	Glu
Leu	Ala	Arg	Pro	Gly	Ala	Ser	Val	Asn	Leu
Ser	Cys	Lys	Ala	Ser	Gly	Tyr	Thr	Phe	Thr
Asn	Tyr	Trp	Met	Gln	Trp	Va1	Lys	Gln	Arg
Pro	Gly	Gln	Gly	Leu	Glu	Trp	Ile	Gly	Ala
Ile	Tyr	Pro	Gly	Asp	Gly	Asp	Thr	Arg	Tyr
Thr	Gln	Lys	Phe	Lys	Gly	Lys	Ala	Thr	Leu
Thr	Ala	Ala	Lys	Ser	Ser	Ser	Thr	Ala	Tyr
Met	Gln	Leu	Ser	Ser	Leu	Ala	Ser	Glu	Asp
Ser	Ala	Val	Tyr	Tyr	Cys	Ala	Arg	Ser	Gly
Tyr	Tyr	Gly	Ser	Phe	Val	Gly	Phe	Ala	Tyr
Trp	Gly	Gln	Gly	Thr	Leu	Val	Thr	Val	Ser
Ala	•							•	

6. DNA and RNA fragments each encoding an immunoglobulin H chain variable region fragment which contains a base sequence encoding a hypervariable region CDR1 having an amino acid sequence selected from

(1) Ser Tyr Trp Met His;
Asp Tyr Tyr Met Asn; and
Asn Tyr Trp Met Gln;

a base sequence encoding a hypervariable region CDR2 having an amino acid sequence selected from

Ala Ile Tyr Pro Gly Asn Ser
Asp Ile Ser Tyr Ser Gln Asn
Phe Lys Asp;
Phe Ile Arg Asn Lys Ala
Asn Leu Tyr Thr Thr Asp
Tyr Ser Ala Ser Val Lys
Gly;
Phe Ile Arg Asn Lys Ala
Asn Tyr Tyr Thr Thr Glu
Tyr Ser Ala Ser Val Lys
Gly; and
Ala Ile Tyr Pro Gly Asp
Gly Asp Thr Arg Tyr Thr
Glu Lys Phe Lys Gly,

#### a base sequence encoding a hypervariable region CDR3 having an amino acid sequence selected from

(3) Glu Glu Tyr Asp Tyr Asp
Thr Leu Asp Tyr;
Asp Arg Gly Gly Arg Asp
Trp Tyr Phe Asp Val;
Asp Gly Phe Leu Arg Asp
Trp Tyr Phe Asp Val; and
Ser Gly Tyr Tyr Gly Ser
Phe Val Gly Phe Ala Tyr.

#### 7. An immunoglobulin H chain variable region fragment having following base sequence

GAG GTT CAG CTC CAG CAG TCT GGG ACT GTG CTG GCA AGG CCT GGG GCT TCA GTG AAG ATG TCC TGC AAG GCT TCG GGC TAC ACC TTT AAC AGC TAC TGG ATG CAC TGG GTA AAA CAG AGG CCT GGA CAG GGT CTG GAA TGG ATT GGC GCG ATT TAT CCT GGA AAT AGT GAT ATT AGC TAC AGC CAG AAC TTT AAG GAC AGG GCC AAA CTG ACT GCC GTC ACA TCC ACC AGC ACT GCC TAC ATG GAA CTC GCG GTC TAC TCT GCG GTC TAT TTC TGT ACA AAA GAG GAA TAT GAT GAT TAC GAC ACC CTG GAC TAC TCG GGT CAA GGG GCC AAA CTG GCG GTC TAC TCT GCG GTC TAC TCT GCG GTC TCA CTCA GCC GTC TCC TCA.

## 8. An immunoglobulin H chain variable region fragm in thaving the following base sequence

GAG GTG AAG CTG GTG GAG TCT GGA GGA GGC
TTG GTA CAG CCT GGG GGT TCT CTC AGA CTC
TCC TGT GCA ACT TCT GGG TTA ACC TTC ACT
GAT TAC TAC ATG AAC TGG GTC CGC CAG CCT
CCA GGA AAG GAA CTT GAA TGG TTG GGT TTT
ATT AGA AAC AAA GCT AAT CTT TAC ACA ACA
GAC TAC AGT GCA TCT GTG AAG GGT CGG TTC
ACC ATC TCC AGA GAT AAT CCC CAA AGC ATC
CTC TAT CTT CAA ATG AAC ACC CTG ACA ACT
GAG GAC AGT GCC ACT TAT TAC TGT GCA AGA
GAT AGG GGG GGG AGG GAC TGG TAC TTC GAT
GTC TGG GGC GCA GGG ACC ACG GTC ACC GTC

## 9. An immunoglobulin H chain variable region fragment having the following base sequence

GAG GTG AAG CTG GTG GAG TCT GGA GGA GGC
TTG GTA CAG CCT GGG GGT TCT CTG AGA CTC
TCC TGT GCA ACT TCT GGG TTC ACC TTC ACT
GAT TAC TAC ATG AAC TGG GTC CGC CAG CCT
CCA GGA AAG GCA CTT GAG TGG TTG GGT TTT
ATT AGA AAC AAA GCT AAT TAT TAC ACA ACA
GAG TAC AGT GCA TCT GTG AAG GGT CGG TTC
ACC ATC TCC AGA GAT AAT TCC CAA AGC ATC
CTC TAT CTT CAA ATG AAC ACC CTG AGA GCT
GAG GAC AGT GCC ACT TAT TAC TGT GCA AGA
GAT GGG TTC CTA CGG GAC TGG TAC TTC GAT
GTC TGG GGC GCA GGG ACC ACG GTC ACC GTC

#### 10. An immunoglobulin H chain variable region fragment having the following base sequence

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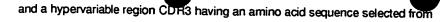
GAG GTT CAG CTC CAG CAG TCT GGG GCT GAA
CTG GCA AGA CCT GGG GCT TCA GTG AAC TTG
TCC TGC AAG GCT TCT GGC TAC ACC TTT ACT
AAC TAC TGG ATG CAG TGG GTA AAA CAG AGG
CCT GGA CAG GGT CTG GAA TGG ATT GGG GCT
ATT TAT CCT GGA GAT GGT GAT ACT AGG TAC
ACT CAG AAG TTC AAG GGC AAG GCC ACA TTG
ACT GCA GCT AAA TCC TCC AGC ACA GCC TAC
ATG CAA CTC AGC AGC TTG GCA TCT GAG GAC
TCT GCG GTC TAT TAC TGT GCA AGA TCG GGC
TAC TAT GGT AGC TCC GTT GGT TTT GCT TAC
TGG GGC CAA GGG ACT CTG GTC ACT GTC TCT
GCG GCC CAA GGG ACT CTG GTC ACT GTC TCT
GCG GCC CAA GGG ACT CTG GTC ACT GTC TCT

#### An immunoglobulin L chain variable region fragment which contains a hypervariable region CDR1 having an amino acid sequence selected from

Gln Leu His Leu Ala Ile Val
Tyr Met His;
Tyr Arg Ala Ser Lys Ser Val
Ser Thr Ser Gly Tyr Ser Tyr
Met His;
Lys Ala Ser Gln Asp Val Asn
Thr Ala Val Ala; and
Lys Ala Ser Gln Asp Val Thr
Thr Asp Val Ala;

#### a hypervariable region CDR2 having an amino acid sequence selected from

(2) Leu Val Ser Asn Leu Glu Ser; Leu Val Ser Asn Leu Asp Ser; and Ser Ala Ser Tyr Arg Tyr Thr,



(3) Gln His Ile Arg Val Ala Tyr
Thr;
Gln His Ile Arg Gly Ala Tyr
Thr;
Gln His Ile Glu Gly Ala Tyr
Thr;
Gln Gln His Tyr Ser Pro Pro
Leu Thr; and
Gln Gln His Tyr Ser Thr Ala
Trp Thr.

## 12. An immunoglobulin L chain variable region fragment having the following amino acid sequence

Asp Ile Val Leu Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Pro Leu Gly Gln Arg Ala Thr Ile Ser Tyr Arg Ala Ser Lys Ser Val Gln Leu His Leu Ala Ile Val Tyr Met His Trp Asn Gln Gln Lys Pro Gly Gln Pro Pro Arg Leu Leu Ile Tyr Leu Val Ser Asn Leu Glu Ser Gly Val Pro Ala Arg Phe Ser Gly Ser Gly Thr Asp Phe Thr Leu Asn Ile His Pro Val Glu Glu Glu Asp Ala Ala Thr Tyr Tyr Cys Gln His Ile Arg Val Ala Tyr Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys .

#### 13. An immunoglobulin L chain variable region fragment having the following amino acid sequence

Asp Ile Val Leu Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Leu Gly Gln Arg Ala Ser Ile Ser Tyr Arg Ala Ser Lys Ser Val Ser Thr Ser Gly Tyr Ser Tyr Met His Trp Asn Gln Gln Lys Pro Gly Gln Pro Pro Arg Leu Leu Ile Tyr Leu Val Ser Asn Leu Glu Ser Gly Val Pro Ala Arg Phe Ser Gly Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Asn Ile His Pro Val Glu Glu Glu Asp Ala Ala Thr Tyr Tyr Cys Gln His Ile Arg Gly Ala Tyr Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys.

#### 14. An immunoglobulin L chain variable region fragment having the following amino acid sequence

Asp Ile Val Leu Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Leu Gly Gln Arg Ala Thr Ile Ser Tyr Arg Ala Ser Lys Ser Val Ser Thr Ser Gly Tyr Ser Tyr Met His Trp Asn Gln Gln Arg Pro Gly Gln Pro Pro Arg Leu Leu Ile Tyr Leu Val Ser Asn Leu Asp Ser Gly Val Pro Ala Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Asn Ile His Pro Val Glu Glu Glu Asp Ala Ala Thr Tyr Tyr Cys Gln His Ile Glu Gly Ala Tyr Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys.

### 15. An immunoglobulin L chain variable region fragm in having the following amino acid sequence

Asp Ile Val Met Thr Gln Ser His Lys Phe Met Ser Thr Ser Val Gly Asp Arg Val Ser Ile Thr Cys Lys Ala Ser Gln Asp Val Asn Thr Ala Val Ala Trp Tyr Gln Gln Lys Pro Gly Gln Ser Pro Lys Leu Leu Leu Tyr Ser Ala Ser Tyr Arg Tyr Thr Gly Val Pro Asp His Phe Thr Gly Ser Gly Ser Gly Thr Asp Phe Thr Phe Thr Ile Ser Gly Val Gln Ala Glu Asp Leu Ala Val Tyr Tyr Cys Gln Gln His Tyr Ser Pro Pro Leu Thr Phe Gly Ala Gly Thr Lys Leu Glu Leu Lys .

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#### 16. An immunoglobulin L chain variable region fragment having the following amino acid sequence

Asp Ile Val Met Thr Gln Ser His Lys Phe Met Ser Thr Ser Val Gly Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Asp Val Thr Thr Asp Val Ala Trp Tyr Gln Gln Lys Pro Arg Gln Ser Pro Lys Leu Leu Ile Tyr Ser Ala Ser Tyr Arg Tyr Thr Gly Val Pro Asp Arg Phe Thr Gly Ser Gly Ser Gly Thr Asp Phe Thr Phe Thr Ile Ser Ser Val Gln Ala Glu Asp Leu Ala Val Tyr Tyr Cys Gln Gln His Tyr Ser Thr Ala Trp Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys

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17. DNA and RNA fragments each encoding an immunoglobulin L chain variable region fragment which contains a bas

	sequence encoding a hypervariable region CDR1 having an amino acid sequence selected from
5	(1) Tyr Arg Ala Ser Lys Ser Val Gln Leu His Leu Ala Ile Val Tyr Met His;
10	Tyr Arg Ala Ser Lys Ser Val Ser Thr Ser Gly Tyr Ser Tyr Met His;
15	Lys Ala Ser Gln Asp Val Asn Thr Ala Val Ala; and Lys Ala Ser Gln Asp Val Thr Thr Asp Val Ala,
20	a base sequence encoding a hypervariable region CDR2 having an amino acid sequence selected from
25	(2) Leu Val Ser Asn Leu Glu Ser;  Leu Val Ser Asn Leu Asp Ser; and  Ser Ala Ser Tyr Arg Tyr Thr,  and a base sequence encoding a hypervariable region CDR3 having an amino acid sequence selected from
<i>30</i>	(3) Gln His Ile Arg Val Ala Tyr Thr;
35	Gln His Ile Arg Gly Ala Tyr Thr; Gln His Ile Glu Gly Ala Tyr Thr;
40	Gln Gln His Tyr Ser Pro Pro Leu Thr; and Gln Gln His Tyr Ser Thr Ala
45	Trp Thr .

## 18. An immunoglobulin L chain variable region fragment having the following base sequence

GAC ATT GTG CTG ACA CAG TCT CCT GCT TCC
TTA GCT GTA TCT CCT CTG GGG CAG AGG GCC
ACC ATC TCA TAC AGG GCC AGC AAA AGT GTG
CAG TTA CAT CTG GCT ATA GTT TAT ATG CAC
TGG AAC CAA CAG AAA CCA GGA CAG CCA CCC
AGA CTC CTC ATC TAT CTT GTA TCC AAC CTA
GAA TCT GGG GTC CCT GCC AGG TTC AGT GGC
AGT GGG TCT GGG ACA GAC TTC ACC CTC AAC
ATC CAT CCT GTG GAG GAG GAG GAT GCT GCA
ACC TAT TAC TGT CAG CAC ATT AGG GTA GCT
TAC ACG TTC GGA GGG GGG ACC AAG CTG GAA
ATA AAA

## 19. An immunoglobulin L chain variable region fragment having the following base sequence

GAC ATT GTG CTG ACA CAG TCT CCT GCT TCC

TTA GCT GTA TCT CTG GGG CAG AGG GCC TCC

ATC TCA TAC AGG GCC AGC AAA AGT GTC AGT

ACA TCT GGC TAT AGT TAT ATG CAC TGG AAC

CAA CAG AAA CCA GGA CAG CCA CCC AGA CTC

CTC ATC TAT CTT GTA TCC AAC CTA GAA TCT

GGG GTC CCT GCC AGG TTC AGT GGC AGT GGG

TCT GGG ACA GAC TTC ACC CTC AAC ATC CAT

CCT GTG GAG GAG GAG GAT GCT GCA ACC TAT

TAC TGT CAG CAC ATT AGG GGA GCT TAC ACG

TTC GGA GGG GGG ACC AAG CTG GAA ATA AAA

#### 20. An immunoglobulin L chain variable region fragment having the following base sequence

GAC ATT GTG CTG ACA CAG TCT CCT GCT TCC
TTA GCT GTA TCT CTG GGG CAG AGG GCC ACC
ATC TCA TAC AGG GCC AGC AAA AGT GTC AGT
ACA TCT GGC TAT AGT TAT ATG CAC TGG AAC
CAA CAG AGA CCA GGA CAG CCA CCC AGA CTC
CTC ATC TAT CTT GTA TCC AAC CTA GAC TCT
GGG GTC CCT GCC AGG TTC AGT GGC AGT GGG
TCT GGG ACA GAC TTC ACC CTC AAC ATC CAT
CCT GTG GAG GAG GAG GAT GCT GCA ACC TAT
TAC TGT CAG CAC ATT GAG GGA GCT TAC ACG
TTC GGA GGG GGG ACC AAG CTG GAA ATA AAA .

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#### 21. An immunoglobulin L chain variable region fragment having the following base sequence

GAC ATT GTG ATG ACC CAG TCT CAC AAA TTC
ATG TCC ACA TCA GTA GGA GAC AGG GTC AGT
ATC ACC TGC AAG GCC AGT CAG GAT GTG AAT
ACT GCT GTA GCC TGG TAT CAA CAG AAA CCA
GGA CAA TCT CCT AAA CTA CTG CTT TAC TCG
GCA TCC TAC CGG TAC ACT GGA GTC CCT GAT
CAC TTC ACT GGC AGT GGA TCT GGG ACG GAT
GAA GAC CTG GCA GTT TAT TAC TGT CAG CAA
CAT TAT AGT CCT CCT CTC ACG TTC GGT GCT
GGG ACC AAG CTG GAA CTG AAA .

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## 22. An immunoglobulin L chain variable region fragment having the following base sequence

GAC	ATT	GTG	ATG	ACA	CAG	TCT	CAC	AAA	TTC
ATG	TCC	ACA	TCA	GTT	GGA	GAC	AGG	GTC	ACC
ATC	ACC	TGC	AAG	GCC	AGT	CAG	GAT	GTG	ACT
ACT	GAT	GTA	GCC	TGG	TAT	CAA	CAG	AAA	CCA
CGA	CAA	TCT	CCT	AAA	CTA	CIG	ATT	TAC	TCG
GCA	TCC	TAT	CGG	TAC	ACT	GGA	GTC	CCT	GAT
CGC	TTC	ACT	GGC	AGT	GGA	TCT	GGG	ACG	GAT
TTC	ACT	TTC	ACC	ATC	AGC	AGT	GTG	CAG	GCT
GAA	GAC	CTG	GCA	GTT	TAT	TAC	TGT	CAG	CAA
CAT	TAT	AGT	ACT	GCG	TGG	ACG	TTC	GGT	GGT
GGC	ACC	AAG	CTG	GAA	ATC	AAA	•		

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23. An Fv region fragment comprising the immunoglobulin H chain variable region fragment according to claim 2 and the immunoglobulin L chain variable region fragment according to claim 12.

24. An Fv region fragment comprising the immunoglobulin H chain variable region fragment according to claim 2 and the immunoglobulin L chain variable region fragment according to claim 13.

25. An Fv region fragment comprising the immunoglobulin H chain variable region fragment according to claim 3 and the immunoglobulin L chain variable region fragment according to claim 14.

26. An Fv region fragment comprising the immunoglobulin H chain variable region fragment according to claim 4 and the immunoglobulin L chain variable region fragment according to claim 15.

27. An Fv region fragment comprising the immunoglobulin H chain variable region fragment according to claim 5 and the immunoglobulin L chain variable region fragment according to claim 16.

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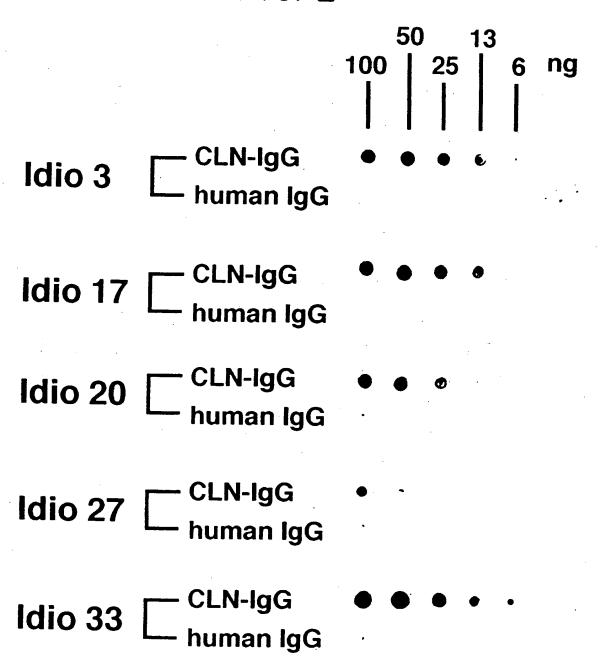
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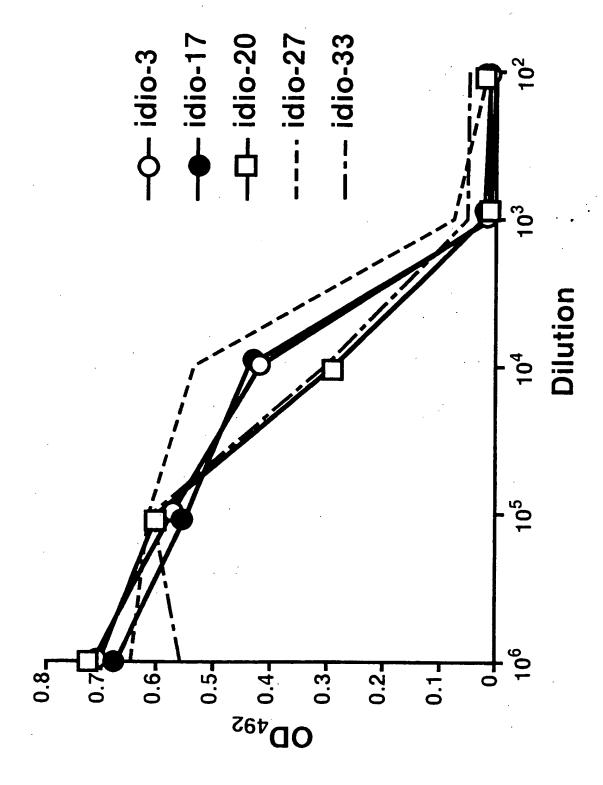
FIG. I

ldio 3	+	λ	K	GЗ	G2b	G2a	9 mere	м	A
Idio 17	4	λ	K	<b>G3</b>	G2b	G2a	G	м	A
ldio 20	+	λ	K	G3	G2b	G2a	Gh	м	A
ldio 27	- Adams	λ	K	G3	G2b	G2a	G1	м	Α
Idio 33	+ 49	λ	κ.	G3	G2b	G2a	G1	м	A

FIG. 2







# FIG. 4

## 3 17 20 27 33

	r					1 21	_ 33
	1	1	GL				
	ĺ	1	l Vai				
	ł	4	Lei		-,-		
	ļ	9	i I Gli				
	1	€	Gli				
		7			· Ser		Ser
		8	1 3				
		9 10					
		11					
		12				Val	Ala
ļ		13					Arg
ı	_	_ 14				Pro	Pro
	F	15		, Gly			Gly
1							Ala
ļ	1	18				Ser Leu	Ser Val
ı		19				Arg	Asn
1		20				Leu	Leu
1		21	Ser	Ser	Ser	Ser	Ser
١		22	Cys			Cys	Cys
١		23	Lys			Ala	Lys
١		24 25	Ala			Thr	Ala
I		26	Ser			Ser Gly	Ser Gly
ı		27	Tyr		Leu	Phe	Tyr
ı		28	Thr		Thr	Thr	Thr
ı		29	Phe		Phe	Phe	Phe
ŀ		30	Asn		Thr	Thr	Thr
ı	S	31	Ser	Ser	Asp	Asp	Asn
I	Ř		Tyr	Tyr Trp	Туг Туг	Tyr Tyr	Tyr
١	1	34	Met	Met	Met	Met	Met
ļ							
1		35	His	His	Asn	Asn	Gln
ı		36	Trp	Trp	Trp	Trp	Trp
١		37	Val	Val	Val	Val	Val
ı		38	Lys	Lys	Arg	Arg	Lys
İ		39	Gln	Gln	Gln	Gln	Gln
ı		40	Arg	Arg	Pro	Pro	Arg
ı		41 42	Gly	Pro Gly	Pro	Pro	Pro
	F	43	Gln	Gln	Gly Lys	Gly Lys	Gly
ļ	F R 2	44	Gly	Gly	Glu	Ala	Gly
ı	2	45	Leu	Leu	Leu	Leu	Leu
l		46	Glu	Glu	Glu	Glu	Glu
l		47	Trp	Trp	Trp	Trp	Trp
H		<u>48</u> 49	Ile Gly	<u> Ile</u> Gly	Gly	Gly	Ile
ı		sø	Ala	Ala	Phe	Phe	Gly
ı		51	Ile	Ile	Ile	Ile	Ile
l		52	Tyr	Tyr	Arg	Arg	Tyr
		SZA	Pro	Pro	Asn	Asn	Pro
l		52B 52C			Lys	Lys	
ŀ		53	Gly	Gly	Ala Asn	Ala Asn	Gly
		54	Asn	Asn	Leu	Tyr	Asp
	Ç	55	Ser	Ser	Tyr	Tyr	Gly
	D	56	Asp	Asp	Thr	Thr	Asp
	Ŗ	57	Ile	Ile	Thr	Thr	Thr
	2	58 59	Ser	Ser	Asp	Glu	Arg
		60	Tyr Ser	Tyr Ser	Tyr Ser	Tyr Ser	Tyr  Thr
		61	Gln	Gln	Ala		Gln
		62	Asn	Asn	Ser		Lys
		63	Phe	Phe	Val	Val	Phe
_		64	Lys	Lys Asp	Lys Gly		Lys
		66	Asp Arg	Asp		Gly Arg	Gly Lys
		67	Ala	Ala	Phe	Phe .	Ala
				_			

68	Lys	Lys	Thr	Thr	Th	_
69	Leu				Le	
70	Thr	Thr			Th	
71	Ala	Ala	Arg		Al	
72	Val					
73	Thr		Asn	Asn	Ly:	
74	Ser				Se	r
75	Thr	Thr		Gln	Sei	r
76	Ser	Ser		Ser	Sei	r
77.	Thr	Thr		Ile	The	r
78	Ala	Ala		Leu	Ald	3
79	Tyr	Tyr		Tyr	Tyı	•
F 80	Met	Met		Leu	Met	
R 81	Glu	Glu		Gln	Gli	
3 8ZA	Leu	Leu	Met	Met	Lei	
82B	Arg	Arg	Asn	Asn	Ser	
82C	Ser	Ser	Thr	Thr	Ser	
83	Leu	Leu	Leu	Leu	Lei	
84	Thr	Thr	Thr	Arg	Alc	
85	Asn	Asn	Thr	Ala	Ser	
86	Glu	Glu	Glu	Glu	Glu	
87	Asp	Asp	Asp	Asp.	Asp	- 1
88	Ser	Ser	Ser	Ser	Ser	
89		Ala	Ala	Ala	Ala	
90	Val	Val	Thr	Thr	Val	
91	Туг	Tyr	Tyr	Tyr	Tyr	
92	Phe	Phe	Туг	Tyr	Tyr	
93	Cys	Cys	Cys	Cys	Cys	
94	Thr  Lys	Thr	Ala	Ala	Ala	
95	Glu	<u>Lys</u> Glu	Arg Asp	Arg	Arg	-
96	Glu	Glu	Arg	Asp	Ser	- 1
97	Tyr	Туг	Gly	Gly Phe	Gly Tyr	1
98	ASP	Asp	Gly	Leu	Typ	ı
•	1	asp	ory	ren	Tyr	ı
	1_					!
99	Tyr	Tyr	Arg	Arg	Gly	l
C 100	ASP	Asp	Asp	Asp	Ser	ı
D 100A	Thr	Thr			Phe	ı
R 1008					Val	ļ
3 100					Gly	ı
1000						١
100E	I					۱
100F						ı
100G						1
100H						ı
100I			Trp	Irp		ı
100)			Tyr	Tyr		I
100K	Leu	Leu	Phe	Phe	Phe	ı
101 102	Asp	Asp	Asp	Asp	Ala	١
102	Tyr	Tyr	Val	Val	Tyr	l
103	Trp	Trp	Trp	Trp	Trp	1
405	Gly	Gly	Gly	Gly	Gly	ĺ
105	Gln	Gln	Ala	Ala	Gln	
F 107	Gly	Gly	Gly	Gly	Gly	ı
R 108	Thr	Thr	Thr	Thr	Thr	l
4 109	Ser	Ser	Thr	Thr	Leu	
110	Val	Val	Val	Val .	Val	
110	Thr	Thr	Thr	Thr	Thr	
112	Val	Val	Val	Val	Val	
113	Ser	Ser	Ser	Ser	Ser	
113	Ser	Ser	Ser	Ser	Ala	

# FIG. 5

3 17 20 27 33

		<u> </u>	1/	20	21	<u> </u>
	1	Asp	Asp	Asp	Asp	Asp
	2	Ile	Ile	Ile	Ile	Ile
	3	Val	Val	Val	Val	Val
	4	Leu	Leu	Leu	Met	Met
	5	Thr	Thr	Thr	Thr	Thr
	6	Gln	Gln	Gln	Gln	Gln
	7	Ser	Ser	Ser	Ser	Ser
	8	Pro	Pro	Pro	His	His
	10	Ala Ser	Ala Ser	Ala Ser	Lys Phe	Lys Phe
	11	Leu	Leu	Leu	Met	Het
F	12	Ala	Ala	Ala	Ser	Ser
R	13	Val	Val	Val	Thr	Thr
ï	14	Ser	Ser	Ser	Ser	Ser
	15	Pro	Leu	Leu	Val	Val
	16	Leu	Gly	Gly.	Gly	Gly
	17	Gly	Gln	Gl'n	Asp	Asp
	18	Gln	Arg	Arg	Arg	Arg
	19	Arg	Ala	Ala	Val	Val
	20	Ala	Ser	Thr	Ser	Thr
•	21	Thr	Ile	Ile	Ile	Ile
	22	Ile	Ser	Ser	Thr	Thr
	23_	Ser			Cys	Cys
	24	Туг	Tyr	Туг	Lys	Lys
	25	Arg	Arg	Arg	Ala	Ala
	26	Ala	Ala	Ala	Ser	Ser
	27	Ser	Ser	Ser	Gln	Gln
	27A	Lys	Lys	Lys		
	27B	Ser	Ser	Ser		
С	27C	Val	Val	Val		
ă	270	Gln	Ser.	Ser Thr		
Ř	27E 27F	Leu	Thr			
1	28	His	Ser	Ser	Asp	Asp
ı		260				
	29	Ala	Gly	Gly	Val	Val
	30	Ile	Tyr	Tyr	Asn	Thr
	31	Val	Ser	Ser	Thr	Thr
	32	Tyr	Tyr	Tyr	Ala	Asp
	33 34	Met	Met	Met His	Val Ala	Val Ala
	35	His Trp	His Trp	Trp	Trp	Trp
	36	Asn	Asn	Asn	Tyr	Tyr
	37	Gln	Gln	Gln	Gln	Gln
•	38	Gln	Gln	Gln	Gln	Gln
_	39	Lys	Lys	Arg	Lys	Lys
F	40	Pro	Pro	Pro	Pro	Pro
R	41	Gly	Gly	Gly	Gly	Arg
2	42	Gln	Gl'n	Gln	Gln	Gln
	43	Pro	Pro	Pro	Ser	Ser
	44	Рго	Pro	Pro	Pro	Pro
	45	Arg	Arg	Arg	Lys	Lys
	46	Leu	Leu	Leu	Leu	Leu
	47	Leu	Leu	Leu	Leu	Leu
	48	Ile	Ile	Ile	Leu	Ile
	49	Tyr	Tyr	Туг	Tyr	Tyr
	50	Leu	Leu	Leu	Ser	Ser
С	51 52	Val	Val	Val Ser	Ala Ser	Ala Ser
Ď	52 53	Ser Asn	Ser Asn	Ser Asn	Ser Tyr	5ег Туг
Ř	53 54	Leu	Leu	Leu	Arg	Arg
2	55	Glu	Glu	Asp	Tyr	Туг
_	56	Ser	Ser	Ser	Thr	Thr
	57	Gly	Gly	Gly	Gly	Gly
	58	Val	Val	Val	Val	Val
	59	Pro	Pro	Pro	Pro	Pro
	60	Ala	Ala	Ala	Asp	Asp
	61	Arg	Arg	Arg	His	Arg
	62	Phe	Phe	Phe	Phe	Phe
	63	Ser	Ser	Ser	Thr	Thr
	64	Gly	Gly	Gly	Gly	Gly

	65	Ser	Ser	Ser	Ser	Ser
	66	Gly	Gly	Gly	Gly	Gly
	67	Gly	Ser	Ser	Ser	Ser
	68	Gly	Gly	Gly	Gly	Gly
	69	Thr	Thr	Thr	Thr	Thr
	70	Asp	Asp	Asp	Asp	Asp
	71	Phe	Phe	Phe	Phe	Phe
	72	Thr	Thr	Thr	Thr	Thr
	73	Leu	Leu	Leu	Phe	Phe
	74	Asn	Asn	Asn	Thr	Thr
	75	Ile	Ile	Ile	Ile	Ile
	76	His	His	His	Ser	Ser
	77	Pro	Pro	Pro	Ser	Ser
· E	78	Val	Val	Val	Val	Val
R	79	Glu	Glu	Glu	Gln	Gln
3	80	Glu	Glu	Glu	Ala	Ala
	81	Glu	Glu	Glu	Glu	Glu
	82	Asp	Asp	Asp	Asp	Asp
	83	Ala	Ala	Ala	Leu	Leu
	84	Ala	Ala	Ala	Ala	Ala
	85	Thr	Thr	Thr	Val	Val
	86	Туг	Туг	Туг	Туг	Tyr
	87	Tyr	Tyr	Tyr	Tyr	Tyr
	88_	Cys	Cys	Cys	Cys	Cys
	89	Gln	Gln	Gln	Gln	Gln
	90	His	His	His	Gln	Gln
	91	Ile Arg	Ile	Ile	His	His
C	92	Arg   Val	Arg Gly	Glu Gly	Tyr Ser	Tyr Ser
CD	93 94	Ala	Ala	Ala	Pro	Thr
Ř			ALG	ALG	Pro	Ala
3	95 95A					ALU
	958					
	95C					
	9SD					
	95E					
	95F					
	96	Tyr	Туг	Tyr	Leu	Trp
	97_	Thr	<u>Thr</u>	<u>Ihr</u>	Thr	Ihc
	98	Phe	Phe	Phe	Phe	Phe
	99	Gly	Gly	Gly	Gly	Gly
F	100	Gly	Gly	Gly	Ala	Gly
R	101	Gly	Gly	Gly	Gly	Gly
4	102	Thr	Thr	Thr	Thr	Thr
7	103	Lys	Lys	Lys	Lys	Lys
	104	Leu	Leu	Leu	Leu	Leu
	105	Glu	Glu	Glu	Glu	Glu
	106	Ile	Ile	Ile	Leu	Ile
	106A		1			
	107	Lys	Lys	Lys	Lys	Lys
Щ		·				



## **EUROPEAN SEARCH REPORT**

Application Number EP 94 11 5683

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···	The present search report has been dr	nwa up for all claims				
	Place of search	Date of completion of the nearch	<u> </u>	Examiner		
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P: interm	ritten disclosure ediate document	& : member of the sa	& : member of the same patent family, corresponding document			



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	Place of search THE HAGUE	Date of completion of the search  16 March 1995	No	examinar oij, F
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